

Fluorescence Assay of Brassinosteroids in Vitazyme — 2023

A study using fluorometric analysis of Vitazyme has confirmed the presence of brassinosteroids in Vitazyme. This involved the chemical reaction between DAPBA (m-dansylaminophenylboronic acid) and brassinosteroids of all types, and then the analysis of these compounds by fluorometric instrumentation.

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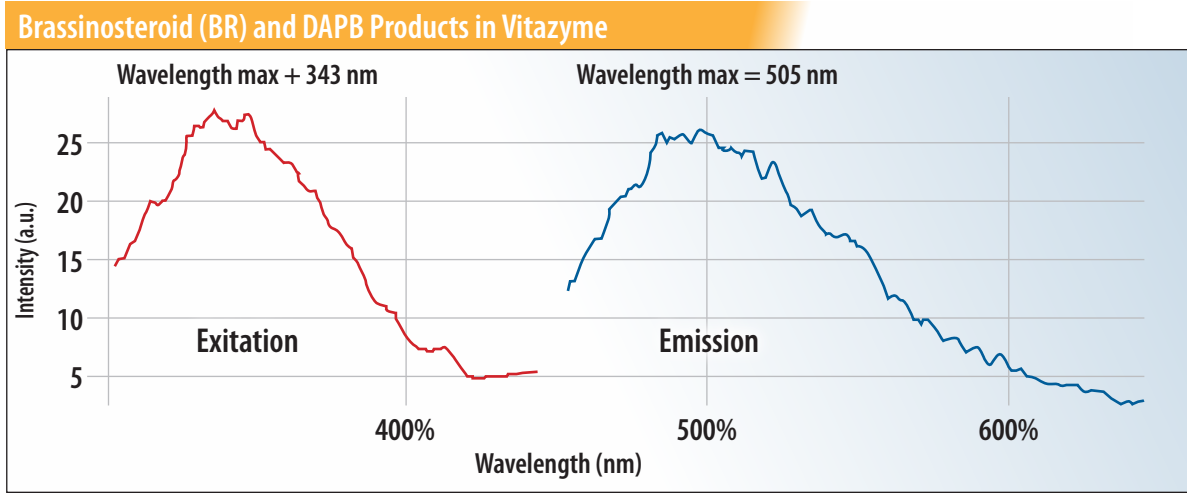
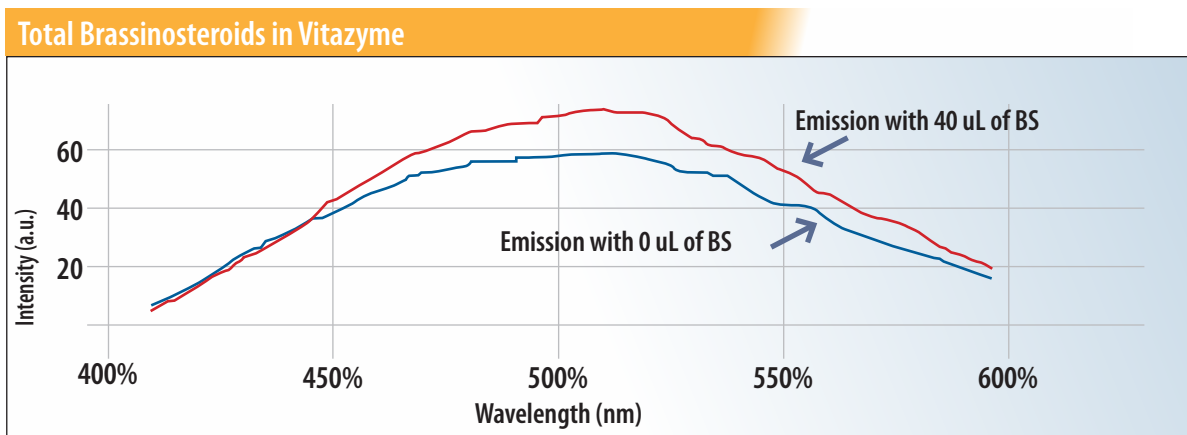


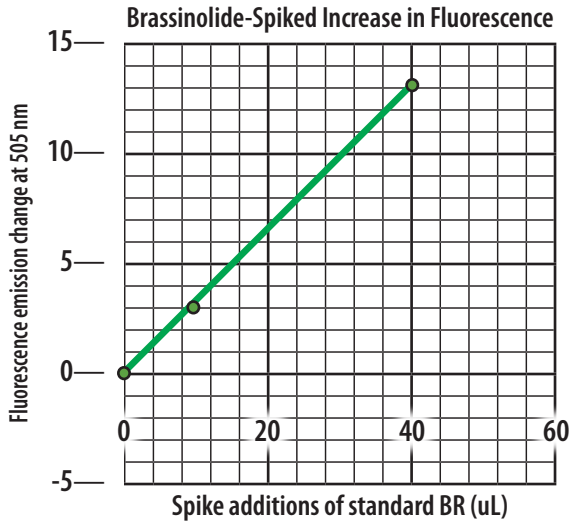
Figure legend: Left trace. Excitation spectrum (produced emission at 505 nm), and right trace emission spectrum (after excitation at 342 nm) of the product of the reaction of 1 mL of Vitazyme with 100 uL of 1mM DABA in 1 mL of phosphate Buffer pH 7.4. The emission wavelength is that expected from a brassinosteroids/DABA reaction product.

Brassinosteroids Spike Reaction



The Vitazyme/DAPBA reaction was spiked with 0, 10, and 40 uL of Brassinolide standard solution (100 uM) The upper trace shows the fluorescence emission scans for 40 uL addition and lower trace for 0 uL addition (10 uL of buffer).

Increase in Fluorescence Intensity at 505 nm After 0, 10, and 40 μL of Brassinolide are Added



Conclusions: These results with fluorometric analysis of Vitazyme confirm the presence of brassinosteroids in the product. The analysis shows the total brassinosteroids, not just brassinolide. The content is $17 \mu\text{M}$ (6ppm) which is less than has been detected by other laboratories using GC/MS analysis. It is expected that different analytic methods will give different results for these very minute quantities, but the presence of the compounds is now confirmed by this analytical method. Further research is ongoing in quantifying brassinosteroids in Vitazyme.