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### **RElative QUantitation Inferred by Evaluating Mixtures (REQUIEM)**

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#### **Running Title**

RElative QUantitation Inferred by Evaluating Mixtures

#### Abstract

Motivated by the lack of easily implementable and generally applicable strategies to increase and assess data accuracy, we devised a novel label-free approach, termed REQUIEM, to address challenges in relative quantitation. For comparing the relative amounts of analytes in two samples, a mixture is prepared from aliquots of the samples, and the samples and the mixture are analyzed in parallel according to the intended workflow. Processing of the resulting data using the REQUIEM algorithm

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Keywords: Relative Quantitation, Precision, Accuracy, Mass Spectrometry, RNA-seq

Abbreviations: ES, embryonic stem (cells); GC-FID, gas chromatography with flame ionization detection; HPAEC-

- PAD, high performance anion exchange chromatography with pulsed amperometric detection; REQUIEM, RElative
- QUantitation Inferred by Evaluating Mixtures; RMSD, root mean square deviation; RPKM, reads per kilobase per
- million; SM, smooth muscle (cells); TPM, transcripts per million; XyG, xyloglucan; XyGO, xyloglucan oligosaccharide. Author Contributions: Study design and direction: WSY. Sugar and glycan sample preparation and data acqui-
- sition: STT. RNA-seq data acquisition and processing: AVN, KWM. REQUIEM data processing and interpretation:

WSY, STT. Statistical analysis: WSY, PS, STT. Software development: WSY.

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