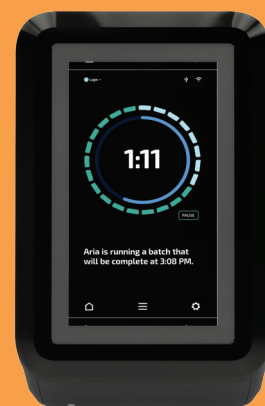


High variations of analyte levels are detected in Terrific broth (TB) media.



BACKGROUND

Terrific broth (TB) is an enriched bacterial media commonly used for high-density cultures of recombinant *E. coli* for protein expression or plasmid purification. Compared to other conventional bacteria media like Lysogeny broth (LB), it has higher concentrations of peptone, yeast extract, and glycerol as a carbon source. Another critical difference between TB and LB is mono and dibasic phosphates in the media, which improve the buffering capacity. TB media is easily acquired premixed in liquid or dry formulations from commercial sources. Although all sources use the same known formulation to mix the media, the exact concentrations of the individual media components may differ due to the different sources of the raw materials. These differences may affect the culture's growth or productivity.

THE EXPERIMENT

All commercially available liquid TB media was tested fresh and handled following the manufacturer's instructions. All samples were diluted 250x before analysis on the REBEL with no additional sample preparation. (Figure 1)

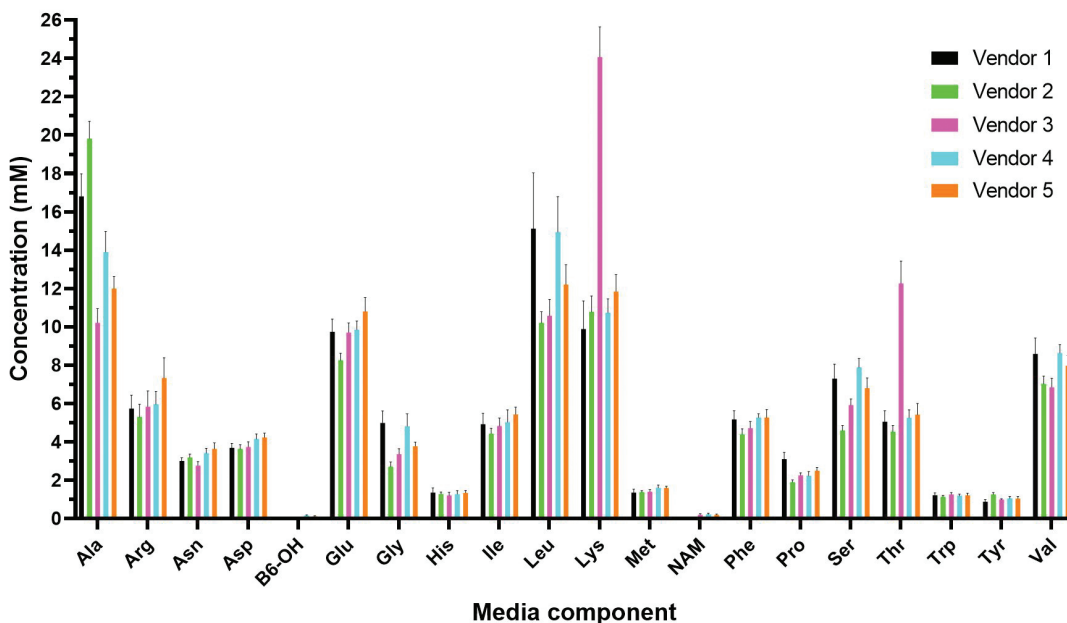


Figure 1: Media component concentrations from Terrific broth diluted by 250x before analysis. Error bars are from the standard deviation of n = 5 replicates.

DISCUSSION

There were several media components with similar concentrations across the five different vendors tested. For instance, the individual concentrations of Arg, Asn, Asp, Glu, His, Ile, Leu, Met, Phe, Trp, Tyr, and Val across all five vendors were all within $\pm 20\%$ of the average of each individual component. For the two B-vitamins detected in this study, B6-OH (pyridoxine) and NAM (nicotinamide), vendors 1, 2, and 3 had no detectable levels of B6-OH, and vendors 1 and 2 had no detected levels of NAM at the dilution level used in this experiment. The most considerable deviations from the average detected levels were observed in the amino acids Ala, Gly, Lys, and Thr. Vendor 2's level of Ala and Gly was 36% higher and 31% lower, respectively, than the average value of the five vendors' media. Vendor 3 had a 79% higher concentration of Lys compared to the average of the other vendors. Finally, vendors 2 and 3 had a 30% lower and 89% higher level, respectively, of Thr compared to the averages. These extreme differences may influence growth or productivities, so a quick media screen before the start of culture is advised. Identifying concentration changes across subsequent runs can help build process knowledge to prevent unexpected deviations.

