



## 2021 ASBC Research Council Grantee

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**Project Title:** *Strategies to prevent growth of pathogens and spoilage organisms in no and low alcohol beers*

### Project Intro:

The no/low alcohol beer (NABLAB) category is a rapidly growing market, with a projected growth rate of 8.28% from 2019-2027 and expected to reach a market value of USD9.26Bn[1]. While much of this growth has been in products from large multi-national brewers, there is sustained and increasing interest from the craft sector. NABLABs are characterised by a high level of nutrients, reduced ethanol, elevated pH, lower hop bitterness and/or milder carbonation[2]. Consequently, the microbial environment associated with NABLABs is significantly different to those of traditional beer types. From a microbial perspective, the NABLAB environment is less hostile due to the absence of stress factors that normally restrict growth and survival. Due to this, NABLABs have been found to be more prone to microbial contamination, and reports of spoilage organisms has dramatically increased in number[2], an issue highlighted by a recent recall by Diageo of their Guinness Draught 0.0% beer[3]. It is documented that sub-lethal stress can induce an adaptive stress tolerance response which increases the survivability of microorganisms[4-6]. Currently a microbiological incident survey for NABLABs does not exist[7], but it has been reported that conventional beer spoilage species grow more rapidly in products where key anti-microbial parameters are compromised[7, 8]. It has also been shown that NABLAB is a viable medium for the growth of *E.coli* 0157:H7 and *Salmonella typhimurium*[9]. Although these organisms can be managed by pasteurisation in bottles and cans, the rise in popularity of kegged NABLABs in the craft market means new strategies for preservation are needed.

### Project Objectives:

To avoid microbial growth in beer, pasteurisation is often utilised. Due to the lower microbiological hurdles present in no and low alcohol beers, these products require a more severe pasteurisation. In recent years kegged low alcohol beers have become more popular, and although large breweries have the ability to pasteurise keg, or use integrity tested sterile filters, it may not be economically viable for smaller breweries to do so. In the latter sector antimicrobial compounds are commonly used to help limit microbial growth and/or destroy pathogenic and spoilage microorganisms [10]. The use of sulphites in low alcohol unpasteurised keg has become standard practice, with a maximum allowable dose of 20 ppm based on US, UK and EU regulations. Other preservatives, both natural and synthetic are available to help with the preservation of beer. The primary aim of this project is to understand the challenges around the microbial stability of unpasteurised low and no alcohol beer, and to investigate the various parameters that can be manipulated to preserve products in the market place. To help simplify this approach we will utilise custom BioLog Phenotype Microarrays (PMs) for the assessment of microbial growth in low and no alcohol beers incubated under a wide range of conditions. These will include

variations in pH, sugar content and the presence of antimicrobials (sulphites, benzoates, sorbates etc.). The PM functions through the reduction of a colourless tetrazolium dye to a purple formazan by electrons from the NADH produced during cellular respiration [11]. The PM platform is flexible, allowing users to construct their own assays using plates, growth media and tetrazolium dyes. Thus, they have proven extremely flexible in terms of the experiments that can be carried out with them [12]. Among other uses, Biolog PM technology has been used in research aimed at understanding and controlling the performance of biotechnological processes, through analysis of microbial metabolism in conditions relevant to industrial fermentations [13-15]. The ultimate aim here is to determine a consistent, economic and practical method for breweries producing low and no alcohol beer in keg, to serve a safe, stable, and quality product to consumers. To do this we will address the following questions: (1) What is the survivability of spoilage and pathogenic microorganisms in no and low alcohol beer? (2) How do environmental conditions (e.g. pH and ethanol) impact on the survivability of these microorganisms? (3) How does the inclusion of antimicrobial preservatives (sulphites, sorbates, benzoates) impact on the survivability of these microorganisms? (4) What is the optimal combination of these factors to produce a safe, stable and quality product in keg?