Assessment of novel yeasts for the production of single cell protein from wasted dates molasses

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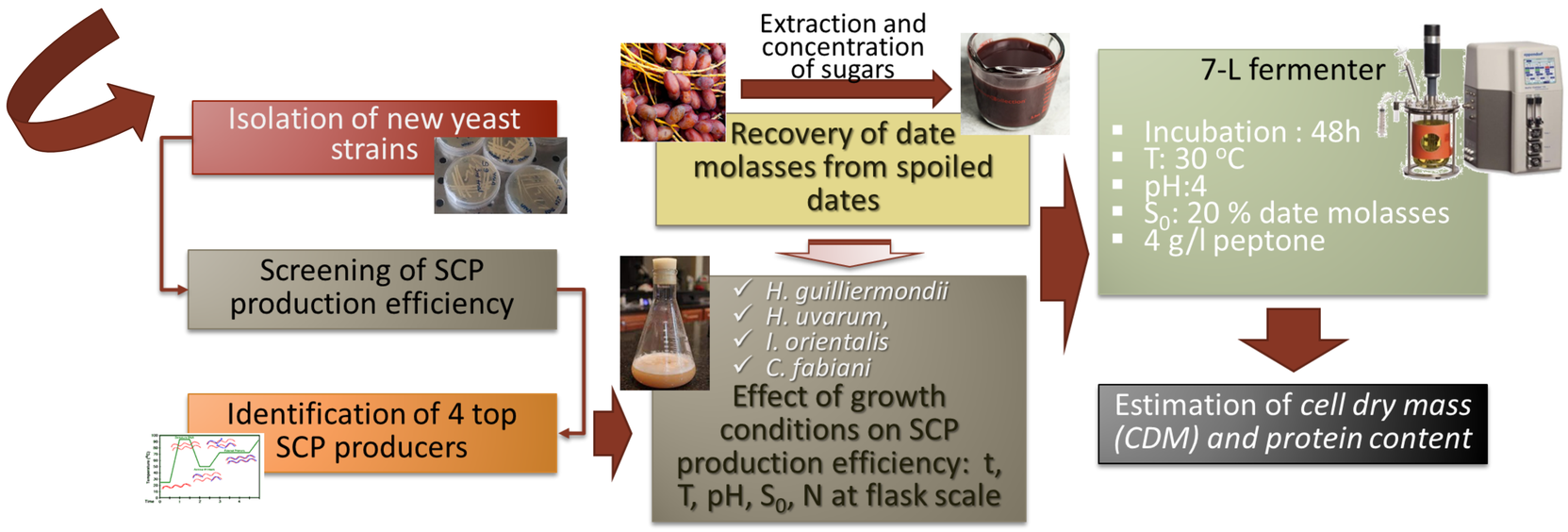
INTRODUCTION: The steadily increasing global population and the subsequent exhaustion of natural resources for animal and human dietary uses have drawn the scientific research during the previous decades to seek for new sources of protein production such as microorganisms. As such, single-cell protein (SCP) production technology arose as a promising alternative to solve the problem of worldwide protein shortage and the focus has shifted to exploit microbes as a potential protein source for humans and as animal feed. Single-cell protein could be produced by many microorganisms; however, fungi and bacteria are preferred because they uptake easily inexpensive waste material but also due to their high growth and protein content. Among them, yeast species are widely accepted as efficient SCP producers, including *Candida utilis, C.robusta, C. tropicalis, Kluyveromyces fragilis, K. lactis, K. marxianus, Sacsharomyces cervesia* etc. Molasses is a sugary by-product, evolving mainly from the processing sugary crops during industrial production of sugar. Date molasses are produced during dates processing, containing high amounts of readily fermentable sugars and other nutrients that can efficiently support yeast growth for SCP production.

Aim: To achieve maximum exploitation of the sugar fraction of wasted dates towards SCP via novel yeast strains exploring the:

- ✓ *long term conservation of date sugars until use*
- ✓ *optimal conditions for SCP production*
- ✓ *scaling up of the process*

Approach & Methodology



Key parameters effecting yeast growth tested :

- ➢ *incubation time : 12, 24, 36, 48, 60, 72, 84, 96 h*
- ➢ *Temperature : 20, 25, 30, 35,40 °C*
- ➢ *initial pH values : 4.0, 5.0, 6.0, 7.0*
- ➢ *substrate concentration : 10 15, 20, 25 and 30%*
- ➢ *nitrogen source : peptone and NH_4Cl*
- ➢ *nitrogen concentration: 0, 1, 2, 3, 4, 5 g/L*

Results

Incubation time, t:
- ✓ Optimum time for maximum production of biomass and protein → 48-60 h,

Temperature, T:
- ✓ Optimum for SCP production for all yeasts : 30 °C
- ✓ Highest biomass production : 1. *I. orientalis* 2. *H. guilliermondii*
- ✓ Highest protein content : *H. guilliermondii*
- ✓ Strong correlation between biomass and protein content (r^2 = 0.901 – 0.976)

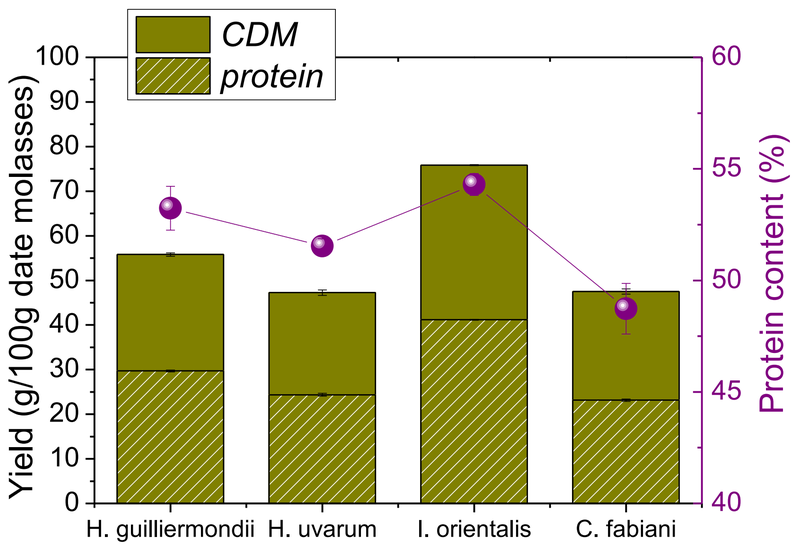
pH :
- ✓ Weak acidity i.e. pH 4-5 optimal for all yeasts

Substrate concentration, S_0 :
- ✓ Optimal 20% for both growth and protein content
- ✓ *I. orientalis* → highest biomass
- ✓ *H. guilliermondii* → highest content of protein

Nitrogen, N:
- ✓ Peptone enhanced both biomass and protein for all yeasts

Effect of temperature, initial pH and substrate concentration on the biomass yield, $Y_{X/molas.}$, expressed as g cell dry mass (CDM) /100 g date molasses) and protein content of yeast biomass , $P_{\%}$, expressed as g protein/100g CDM) . Values are means of 3 replicates ± standard error. Values for the same parameter tested in the same with the same letter are not significant ($P < 0.001$)

		H. guilliermondii		*H. uvarum*		*I. orientalis*		*C. fabiani*	
		$Y_{X/molas.}$	$P_{\%}$	$Y_{X/molas.}$	$P_{\%}$	$Y_{X/molas.}$	$P_{\%}$	$Y_{X/molas.}$	$P_{\%}$
Temperature	20 °C	24.5±0.98a	32.5±0.90a	23.9±0.91b	30.4±0.59b	39.0±0.29b	35.6±0.43b	20.4±1.07b	30.4±0.59b
	25 °C	47.9±0.72c	48.5±1.07bc	40.6±0.75d	43.4±0.59c	41.4±0.12c	45.1±0.87c	33.3±0.96c	43.4±0.59c
	30 °C	48.2±0.88c	51.5±0.86c	46.2±0.33e	44.2±0.88c	51.0±0.58d	50.8±1.10d	37.5±0.58d	44.2±0.88c
	35 °C	42.7±0.39b	46.5±0.65b	38.0±0.06c	43.2±0.98c	42.2±0.12c	45.5±0.58c	30.1±1.10c	45.2±0.65c
	40 °C	27.5±0.97a	29.5±1.56a	4.9±0.33a	15.9±2.27a	35.6±0.17a	32.5±0.0a	13.6±0.81a	21.5±0.58a
pH	4	53.0±0.12d	51.7±1.12c	48.1±0.81c	46.3±1.11c	54.2±0.12c	50.1±1.11c	43.7±0.79d	46.9±0.81c
	5	48.2±0.17c	50.7±1.12c	45.7±0.77c	48.5±0.58c	49.4±0.06	48.2±1.20c	40.7±0.39c	45.3±0.39c
	6	42.6±0.12b	41.2±0.88b	41.4±0.76b	40.4±0.59b	36.5±1.33bc	37.1±1.27b	35.1±0.81b	37.1±1.27b
	7	25.7±1.47a	36.1±0.43a	30.4±0.76a	34.7±1.18a	20.2±2.85a	30.8±0.20a	19.8±0.88a	26.8±0.88a
Substrate concentration	10%	45.8±0.12ab	45.4±1.74a	42.4±0.67c	45.3±3.47bc	41.9±0.78b	48.2±0.88bc	35.8±0.38a	45.5±1.73bc
	15%	47.8±0.17b	46.0±0.27a	43.6±0.81c	45.9±0.63bc	47.5±0.58c	52.0±0.40bc	40.7±0.29b	49.7±20.8d
	20%	58.5±3.64c	51.8±0.25b	50.1±0.57d	48.5±0.61d	63.5±1.18d	48.4±1.41d	46.2±0.30c	46.2±1.77bc
	25%	43.2±0.23ab	44.3±0.92a	39.8±0.59b	40.5±0.33ab	44.0±1.73bc	44.7±1.18ab	42.2±1.17b	41.3±0.67ab
	30%	40.4±0.06a	41.5±1.0a	35.4±0.22a	37.5±0.53a	37.2±1.18a	42.9±1.23a	35.5±0.13a	37.2±1.18a



- Scale up experiments led to higher yields compared to flask experiments
- All yeast stains effectively consumed all available sugars reaching maximum growth and productivity
- *I. orientalis* exhibited the highest yield : 75.84 ± 0.14 g CDM/100 g molasses with protein content 54.3 ± 0.49, corresponding to a protein yield of 41.15 ± 0.07 g protein/100 g molasses

Conclusions

The study introduces four new effective yeasts and a new economic substrate for production of large amount of SCP.

This substrate represents an efficient substrate for yeasts growth when amended with extra nitrogen for increased productivity.

The study indicates that the new yeasts are promising candidates SCP-producer for industrial and commercial production as animal feed

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