CHAPTER VII

PREPARATION OF RAW MATERIAL

7.1 Washing yeast cells

7.1.1 Apparatus

Bowl centrifuge in twin tubs washing machine (Sharp, 1243 rpm), with a cotton cloth filter bag as inner lining.

7.1.2 Reagents

7.1.2.1 Sodium carbonate solution 2%

7.1.2.2 Sodium hydroxide solution 30%

7.1.3 Procedure

7.1.3.1 Alkaline washing

i) Harvested yeast cells from fermentors,

ii) Separated beer and yeast cells by centrifugation in bowl centrifuge at 1243 rpm, collected yeast cells.

iii) Added 2% sodium carbonate solution to yeast cells at 1:1 (w/w).

iv) Mixed to disperse the yeast cells.

v) Adjusted yeast slurries to pH 10 by 30% sodium hydroxide.

vi) Separated beer and yeast cells by centrifugation in bowl centrifuge at 1243 rpm, collected yeast cells.

vii) Added water to yeast cells at 1:1 (w/w).

viii) Mixed to disperse the yeast cells.

ix) Separated water and yeast cells by centrifugation in bowl centrifuge at 1243 rpm, collected yeast cells.

x) Did the same process from 7.1.3.1 vii to 7.1.3.1 ix till the pH of yeast slurries were neutral.

7.1.3.2 Distilled water washing

Did the same process as 7.1.3.1 except with addition of distilled water in stead of sodium carbonate solution for 3 times at similar ratio.

7.2 Proximate analysis of yeast cells

Yeast cells were analysed for protein (European Brewery Convention, 1975), moisture content (Clerck, 1958).

7.2.1 Protein (European Brewery Convention, 1975)

7.2.1.1 Apparatus

7.2.1.1.1 Digestion unit (BUCHI, 425)

7.2.1.1.2 Distillation unit (BUCHI, 322)

7.2.1.1.3 Fume cupboard

7.2.1.2 Reagents

7.2.1.2.1 Sulphuric acid 98%

7.2.1.2.2 Selenium mixture consists of a powder mixture of potassium sulphate, copper sulphate and selenium in the proportion 100:6:1 (w/w/w)

7.2.1.2.3 Sodium hydroxide solution 32% (w/v)

7.2.1.2.4 Boric acid solution 2% (w/v) in water

7.2.1.2.5 Standard hydrochloric acid, 0.1 N.

7.2.1.2.6 Mixed indicator : bromcresol green, p-nitrophenol, ponceau 4R, 0.1 N. sodium hydroxide, alcohol 96%, distilled water

7.2.1.3 Procedure

i) Weigh 2.0 gm of sample.

ii) Weigh 6 gm of selenium mixture (7.2.1.2.2).

iii) Mix i and ii in macro Kjeldahl digestive tube. Add 20 ml of concentrated sulphuric acid (7.2.1.2.1) and digest in digesting

unit (7.2.1.1.1) and digest until a clear solution is obtained (about 1 hour).

iv) Connect the clear solution to distillation unit (7.2.1.1.2) and diluted with 100 ml distilled water and set up the distillation unit so that the distillate outlet tube is immersed below the liquid level of 50 ml of 2% boric acid (7.2.1.2.4).

v) Distill with 90 ml of 32% sodium hydroxide (7.2.1.2.3) until the distillate in a receiving flask attained a total volume of about 200 ml.

vi) Titrate the distillate with 0.1 N. hydrochloric acid (7.2.1.2.5) using mixed indicator (7.2.1.2.6) as indicator.

vii) Carry out a blank determination on the reagents mentioned above but without sample.

7.2.1.4 Calculation

The percentage of nitrogen on dry basis, $N = \frac{T \times 14}{W \times D}$

when N = Percentage of nitrogen on dry basis.

T = Acid titration value, with the blank substracted and corrected to exact tenth normality (ml).

W = Weight in gm of sample taken.

D = Percentage of dry matter.

Protein is calculated from the amount of nitrogen multiplied by 6.25

7.2.2 Total ash (Indian standard, 1973)

7.2.2.1 Apparatus

7.2.2.1.1 Porcelain dish

7.2.2.1.2 Electric muffle furnance

7.2.2.1.3 Desiccator

7.2.2.2 Procedure

i) Ignite porcelain crucible, cool in the desiccator and weigh.

ii) Weigh 3 gm of yeast slurries in the crucible to the fourth decimal place .

iii) Transfer the crucible and its content to the muffle furnance at 550 C and burn to a white ash .

iv) Transfer the crucible and its content to the desiccator, allow to cool and weigh .

v) Return the crucible to the muffle furnance and ignite again for each 30 minutes until loss of weight between succesively weighing do not exceed 1 mg .

7.2.2.3 Calculation

$$\text{Percentage total ash (based on wet weight)} = \frac{D}{W} \times 100$$

when W = Weight in gm of sample taken before ignition .

D = Weight in gm of dry ash after ignition .

7.2.3 Moisture content (Clerck, 1958)

7.2.3.1 Apparatus

7.2.3.1.1 Electric oven (Kottermann)

7.2.3.1.1 Desiccator

7.2.3.2 Reagent

7.2.3.1.1 Ethanol 95%

7.2.3.1.2 Sand (acid washed sea sand)

7.2.3.3 Procedure

i) Mix the contents of yeast to a uniform consistency .

ii) Place 5 gm of dry sand in the weighing dish .

iii) Place the dish plus sand on the balance pan, together with the stirring rod .

iv) Transfer the well mixed liquid yeast to the weighing dish, cover and weigh to the nearest 0.001 gm .

v) Remove the cover and add 5 ml of ethanol and mix throughly by means of the stirring rod.

vi) Drop the stirring rod into the weighing dish and dry the whole for 3 hours at 105° C

vii) Cover the dish and transfer the dish to desiccator, allow to cool and weigh

7.2.3.4 Calculation

$$\text{Percentage Total solid} = \frac{D}{W} \times 100$$

when W = Weight in gm of the slurry aliquot before drying

D = Weight in gm of the slurry after drying

Percentage Moisture content = 100—% Total solid

7.3 Results

7.3.1 Protein

Analytical results for protein content of spent yeast cells from fermentors of Boon-Rawd Brewery Co., Ltd. between May to October 1984 are shown in Figure 7-1.

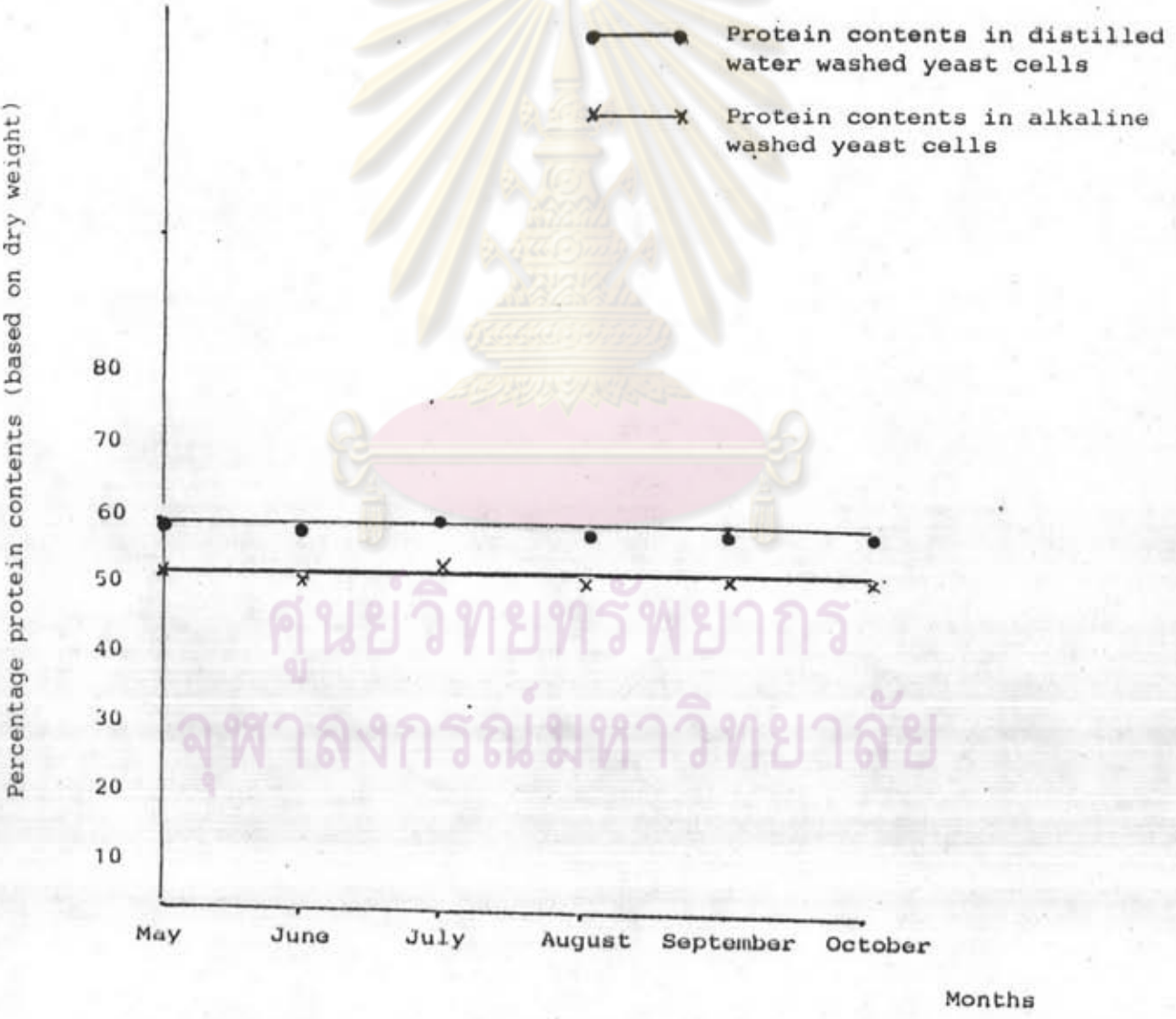

Figure 7-1 Protein contents of locally available brewer's yeast

7.3.2 Composition of spent yeast

Analytical results for ash and moisture contents of washed yeast cells are shown in Table 7-1.

Table 7-1 Chemical composition of spent yeast (expressed in percentage)

Components	Spent yeast
Total ash (based on wet weight)	1.90 ± 0.02
Moisture content	76.33 ± 0.22

7.4 Discussion

7.4.1 Protein

Protein content of alkaline washed yeasts was lower than that of distilled water washed yeasts. Most cytoplasmic materials except cell wall were dissolved when treated yeast cells with alkali (Perlman, 1969). The protein content of each, however, was invariably constant over the period of investigation.

7.4.2 Ash

Ash content (based on dry weight) of spent yeast was similar to published content of spent yeast.