

## OCCURRENCE OF YEASTS AND MOLDS IN HARD CHEESE (ROMI)

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### SUMMARY

Fifty random samples of hard cheese (Romi) were collected from different dairy shops, groceries and supermarkets in Zagazig City Egypt, to be examined mycologically. Yeasts could be detected in 70% of hard cheese samples, with a mean value of  $2.2 \times 10^4 \pm 0.8 \times 10^4$  g. While molds were isolated from 86% of examined hard cheese samples, with a mean value of  $1.3 \times 10^3 \pm 0.4 \times 10^3$ /g. The isolated yeasts were *Torulopsis candida* (22%), *Tor. holmii* (10%), *Tor. globrata* (6%), *Saccharomyces bailii* (20%), *Sac. cerevisiae* (8%), *Candida castellii* (12%), *C. lipolytica* (10%), *Debaryomyces hansenii* (10%), *Kluyveromyces marxianus* (6%), *Rhodotorula pallida* (4%) and *Geotrichum candidum* (4%), while the mold isolates were *Penicillium commune* (24%), *P. verrucosum* (10%), *P. solitum* (8%), *P. crustosum* (6%), *Aspergillus niger* (20%), aflatoxigenic strains of *Asp. flavus* (2%), non-aflatoxigenic strains of *Asp. flavus*

(10%), *Asp. versicolor* (8%), *Cladosporium werneckii* (8%) and *Rhizopus* spp. (6%). The economic and public health importance of isolated fungi as well as the control measures for improving the quality of the product were discussed.

### INTRODUCTION

Cheese provides a valuable contribution to the human diet. It contains a good source of high quality protein, fat soluble vitamins and minerals (Renner, 1983).

Hard cheese has a low water activity, low pH and a higher salt content, so it is considered an excellent medium for growth of many species of fungi (Foster et al., 1983).

Yeast and mold are widely distributed in the environment and can enter the foods through inadequately sanitized equipment or as air-borne,

contaminants, to produce undesirable odour, flavour, discolouration and gas formation (Ray, 1996).

The growth of toxigenic fungi in cheese depends on the temperature, humidity, water activity, gas atmospheres, pH and resistance to preservatives (Hocking, 1997 and Serrano et al., 1997).

Cases of foodborne illness were reported by Todd (1985) due to consumption of dairy products contaminated by yeasts and molds. Also, some species of fungi may cause various diseases in man (Robinson, 1990).

The present investigation was planned to detect the fungal contamination of hard cheese (Romi) in Zagazig City, Egypt.

## MATERIALS AND METHODS

### Sampling:

Fifty random samples of hard cheese (Romi) were collected from different dairy shops, groceries and supermarkets in Zagazig City, Egypt. The collected samples were transferred directly to the laboratory with a minimum of delay to be examined mycologically.

### Preparation of samples:

Each sample was prepared according to the

method recommended by A.P.H.A. (1985).

### Enumeration, isolation and identification of fungi:

One ml. from each serial dilution was plated on duplicate plates of malt extract agar acidified with 10% lactic acid solution and incubated at 25°C for 5 days. The average number of colonies/g. was calculated. The suspected colonies were picked up, streaked onto malt extract agar and incubated at 25°C for 5 days. The pure yeast isolates were subjected for identification according to Looder and Kreger Van-Rij (1970). While, the isolated molds were identified according to the techniques adopted by Raper and Fennell (1965) and Samson et al. (1981).

### Screening for aflatoxin producing strains of *Aspergillus flavus*:

The isolates of *Asp. flavus* were inoculated at the center of cocount agar medium (Davis et al., 1987) and incubated at 28-30°C for at least 2 days, then the plates were inspected daily for the presence of blue fluorescent zone around the colonies on exposure to long wave U.V. light (365nm).

## RESULTS

Table (1): Statistical analytical results of total yeast and mold counts/g. of the examined hard cheese (Romi) samples.

Counts	No. of examined samples	Positive samples		Count/g.			
		No.	%	Min.	Max.	Mean	S.E.M.±
Yeast count	50	35	70	60	1.7X10 <sup>5</sup>	2.2X10 <sup>4</sup>	0.8X10 <sup>4</sup>
Mold count	50	43	86	30	3.6X10 <sup>4</sup>	1.3X10 <sup>3</sup>	0.4X10 <sup>3</sup>

Table (2): Frequency distribution of positive hard cheese (Romi) samples based on their yeast and mold count/g.

Intervals		Frequency			
		Yeast		Mold	
		No. of samples	%	No. of samples	%
10	10 <sup>2</sup>	2	5.7	5	11.6
10 <sup>2</sup>	10 <sup>3</sup>	7	20.0	12	28.0
10 <sup>3</sup>	10 <sup>4</sup>	11	31.4	21	48.8
10 <sup>4</sup>	10 <sup>5</sup>	10	28.6	5	11.6
10 <sup>5</sup>	10 <sup>6</sup>	5	14.3	--	--
<b>Total</b>		<b>35</b>	<b>100.0</b>	<b>43</b>	<b>100.0</b>

Table (3): Incidence of yeasts isolated from the examined hard cheese (Romi) samples .

Isolates	No. of samples	%
Torulopsis candida	11	22
Tor. holmii	5	10
Tor. glabrata	3	6
Saccharomyces bailii	10	20
Sac. cerevisiae	4	8
Candida castelli	6	12
C. lipolytica	5	10
Debaryomyces hansenii	5	10
Kluyveromyces marxianus	3	6
Rhodotorula pallida	2	4
Geotrichum candidum	2	4



Table (4): Incidence of molds from the examined hard cheese (Romi) samples .

Isolates	No. of samples	%
<i>Pencillium commune</i>	12	24
<i>P. verrucosum</i>	5	10
<i>P. solitum</i>	4	8
<i>P. crustosum</i>	3	6
<i>Aspergillus niger</i>	10	20
<i>Asp. flavus</i> :	6	12
- Aflatoxigenic strains	1	2
- non -aflatoxigenic strains	5	10
<i>Asp. versicolor</i>	4	8
<i>Cladosporium werneckii</i>	6	12
<i>Rhizopus spp.</i>	3	5

## DISCUSSION

Results recorded in Table (1) revealed that yeasts were present in 70% of the examined hard cheese (Romi) samples. The total yeast count/g. ranged from 60 to  $1.7 \times 10^5$ , with a mean value of  $2.2 \times 10^4 \pm 0.8 \times 10^4$ . The highest frequency distribution (60%) lies within the range  $10^3 - 10^5$  (Table, 2).

Nearly similar counts were reported by El-Essawy et al. (1984) and Mansour (1985). Comparatively higher results were reported by Eliskases-Lechner (1996) and Welthagen and Viljoen (1996). While lower findings were recorded by Abouzeid et al. (1996).

Results reported in Table (1) showed that 86% of the examined hard cheese (Romi) samples contained molds, with counts/g, ranged from

30 to  $3.6 \times 10^4$ , with an average  $1.3 \times 10^3 \pm 0.4 \times 10^3$ . Most of samples (76.8%) lies within the range of  $10^2 - 10^4$  (Table, 2)

Nearly similar results were reported by Abouzeid et al. (1996). While, higher counts were reported by El-Essawy et al. (1984) and Mansour (1985).

*Torulopsis candida*, *Tor. holmii*, *Tor. glabrata*, *Saccharomyces bailii*, *Sac. cerevisiae*, *Candida castellii*, *C. lipolytica*, *Debaryomyces hansenii*, *Kluyveromyces marxianus*, *Rhodotorula pallida* and *Geotrichum candidum* could be isolated from the examined hard cheese (Romi) samples in percentages ranged from 4% to 22% (Table, 3).

Most of these yeast isolates were detected in hard cheese by Mansour (1985), Fleet and Main (1987), Lund et al. (1995), Abouzeid et al. (1996), Eliskases-Lechner (1996) and Welthagen and

Vet. Med. J., Giza. Vol. 48, No. 4 (2000)

Vijayan (1996).

*Penicillium commune* and *Aspergillus niger* were the predominated molds isolated from the examined hard cheese samples, while *Penicillium verrucosum*, *P. solitum*, *P. crustosum*, aflatoxin producing strains of *Asp. flavus*, non-aflatoxin producing strains of *Asp. flavus*, *Asp. versicolor*, *Cladosporium werneckii* and *Rhizopus* spp. could be isolated at varying percentages ranged from 6% to 24% (Table,4).

Similar Species of mold were isolated from hard cheese by El-Essawy et al. (1984), Mansour (1985), Lund et al. (1995), Abouzeid et al. (1996) and Serrano et al. (1997)

Yeasts may cause cheese spoilage by breaking down its components and liberating different acids and gas with subsequent change of its colour and flavour. Moreover, mold growth on cheese causes economic losses from discoloration, poor appearance and off flavours. In addition, some molds are capable of producing toxic metabolites known as mycotoxins such as aflatoxins which are known carcinogens (Pitt and Hocking, 1985).

Lack and Remmert (1980) reported that the surface treatment of hard cheese with aqueous solutions of potassium sorbate act as a fungistat for cheese. Moreover, Bullerman et al. (1984) reported that antimycotic agents ascorbic acid, potassium sorbate, propionic acid and

Vet. Med. J. Giza. Vol. 48. No. 4 (2000)

propionates, in addition to lower R.H. and moisture content and low temperature can be used to control the mold growth and mycotoxin production.

In conclusion, proper sanitation and hygiene during processing, handling, storage and distribution of hard cheese are important to minimize the fungal contamination of cheese and to safeguard the consumers from infection.

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