Vet.Med.J.,Giza. Vol.42,No.1.(1994):19-22.

DETECTION OF FORMALIN IN RAS CHEESE DURING THE RIPENING PERIOD BY DIFFERENT METHODS

BY

SALEM, S.A.*; A.M. NAZEM** and M.I. EL-NAGGAR***

* Department of Dairy Sci. and Tech., Faculty of Agriculture, Alex. Univ., Egypt..
** Department of Foof Hygiene, Faculty of Veterinary Medicine, Alex. Univ., Egypt.
*** Department of Forensic Medicine and Toxicology, Faculty of Medicine, Alex. Univ. Egypt.

Received: 23/11/1993.

SUMMARY

One hundred and twenty samples of fresh Ras cheese collected from different localities at El-Behera Governorate were examined for the presence of formalin by Hehner's, flame and chromotropic acid tests, then 10% of the positive samples (seven) were stored and tested during ripening period up to 90 days.

Most of cheese samples (58.33%) contained formalin detected by flame and chromotropic acid tests, but 41.67% of the tested samples were positive by Hehner's method.

The chromotropic acid test is accurate, rapid, simple, highly specific, sensitive, inexpensive, easy and quantitative method for detection of formalin especially during ripening period and with low concentrations in routine controls.

There were great variations in formalin content in different samples depend upon the cheese maker. Also, the quantities of formalin in tested samples of Ras cheese were not changed during ripening, whereas it revealed dangerous and highly toxic effect due to daily consumption of the cheese and at a long continuous intake of formalin it migh be harmful. It can be recommended that formalin must not be added to the milk used in manufacturing Ras cheese at any level

INTRODUCTION

Ras cheese is the most popular hard cheese in Egypt and most mediterranean countries. it is made from raw or pasteurized cow's milk or a mixture of buffaloe's and cow's milk. The cheese is consumed after ripening period of at least three months.

Preservatives are illegal in some countries. In other countries, it is believed that they should be used in cases of emergency rather than let the milk spoil, but that they should never be regarded as a substitute for hygienic milk handling (Berg, 1988). Although formalin is hazardous to human health many cheese makers in some countries, as Italy, add formaldehyde to Grana cheese and other Italian hard cheeses because it has a synergistic effect against late blowing by inhibiting propionic acid bacteria (Bottazzi et al., 1984). Also, many cheese makers in Egypt illegally add a small amount of formalin to milk to improve its keeping quality according to the season of milk production. The cheese makers usually add different quantities of formalin depending upon the quality of milk and they believe that formalin inhibits the growth of undesirable bacteria which cause defects and objectionable flavoures in cheese made from raw milk, without harming the quality of cheese (Abdel-Mottaleb et al., 1968), while Mahmoud et al. (1985) found that low concentration of formalin could not be added to cheese milk to inhibit undesirable organisms without also, inhibiting desirable species.

Small amounts of formaldehyde in milk, one in 200,000 parts, can be easily detected (Atherton and Neulander, 1977), while El-Gendy et al. (1980) found that formalin could not be detected

easily after 2 to 4 days in cheese made from milk containing 0.002% or 0.008% of the chemical, whereas when cheese was made from milk with 0.02 to 0.1% formalin, the chemical was detectable for up to 8 weeks. Another method was applied for detecting formalin in cheese by Resmini et al. (1988) by estimating Spinacine which is formed in cheese from the reaction of formaldehyde with the His_{106} residue residue of 2-casein. Also, Restani et al. (1989) applied an isoelectric focusing method to quantify the amount of formaldehyde in cheese by calculating the amount of protein associated with this band.

For these reasons, this work was conducted to study the incidence of formalin in Ras cheese with some screening simple methods of formalin detection in El-Behera Governorate and estimation of its quantities during the ripening period of cheese up to three months by some different methods.

MATERIALS AND METHODS

Cheese samples:

One hundred and twenty random samples of fresh Ras cheese (after pressing) were collected from different localities in El-Behera Governorate. Seven (10%) out of the samples positive for formaldehyde were stored up to 90 days at 12-15°C and 85% relative humidity, then the samples were tested every 30 days for the presence of formalin.

Formaldehyde tests:

1. Hehner's test: according to Ling (1963) as follows: Mix 2 ml of distillate with 2 ml of extracted solution from 2g of cheese in a test-tube. Pour sulphuric acid (90 per cent containign a trace of ferric chloride) down the side of the tube. In the presence of formaldehyde a violet zone forms at the junction of the layers. The violet coloration does not appear usually when relatively large quantities of formaldehyde are present.

2. Flame test: according to Man and Saunders (1946) as follows: warming together carefully 2g of sample and 1 ml of concn. sulfuric acid. Identify the carbon monoxide by igniting the gas evolved and observing the pale blue flame travel down the esttuble.

3. Chromotropic acid test: A purple colour which appears after reaction with chromotropic acid and formic acid in the presence of sulfuric acid indicates the present of formaldehyde in protein free filtrate by trichloroacetic acid according to Clarke (1978). he optical density of the colour at 550 nm was taken after calibration of absorbance with different concentrations of formalin. The concentration of formalin can be calculated in each sample by standard curve.

RESULTS AND DISCUSSION

Table (1) shows that 50.33% of the tested samples were positive for formalin test by flame or chromotropic acid method, while 41.67% out of the examined samples were positive to Hehner's method. It was obvious that most of the cheese makers used formalin as a preservative in cheese manufacturing at this area, therefore, danger is expected by consumption of this cheese due to the high toxicity of formalin even in small amounts.

From the results obtained and recorded it is obvious that the chromotropic acid test is more accurate than the flame or Hehner's test because it is quntitive test, it is easy in application if compared with flame method which needs high accuracy.

Table (1): Incidence of formalin in examined samples of fresh Ras cheese.

Method	No. of examined samples	Positive samples*	
		No.	%
Hehner's	120	50	41.67
Flame	120	70	58.33
Chromotropic acid	120	70	58.33

* The positive samples were the same in all examined samples by different methods.

Table (2) shows the determination of formaldehyde in 10% of the positive Ras cheese samples during ripening period up to 90 days, by different methods.

From the results obtained, it is concluded that the Hehner's test was not available for detection of formalin in ripened cheese. It was clear that this method was not esact during ripening period or th detection after 30, 60 and 90 days especially with low concentration. The flame method gave better results as compared with Hehner's method with all concentrations of formalin during ripening period.

Table (2): Variability of different methods for formalin detection in Ras cheese during ripening period.

Serial No.	Storage period (days)	Hehner's method	Flame Method	Chromotropic acid method (ppm)
1	0	+	+	6.30
	30	-	+	6.00
	60	-	+	6.10
	90	-	+	6.00
2	0	+	+	3.55
	30	-	+	3.60
	60	-	+	3.66
	90	-	+	3.60
3	0	+	+	3.35
	30	-	+	6.33
	60	-	+	6.50
	90	-	+	6.55
4	0	+	+	1.87
	30	-	+	1.92
	60	-	+	1.95
	90	-	+	1.95
5	0	+	+	25.00
	30	+	+	25.00
	60	+	+	24.80
	90	+	+	24.70
6	0	+	+	15.50
	30	+	+	15.70
	60	+	+	15.75
	90	-	+	15.02
7	0	+	+	4.42
	30	-	+	4.85
	60	-	+	5.00
	90	-	+	0.05

The chromatropic acid test proved to be the most preferable and exact method especially during the ripening period of the cheese, therefore, it can be recommended to use this method for the detection and identification of formalin in ripened cheese at all concentrations as compared with Hehner's method.

From Table (2), it could be concluded also that the variations between quantities of formalin in the samples were very high.

It should be borne in mind that the results obtained tend to be rather lower than the amount originally added due to decomposition after incorporation into the food and to low recoveries in distillates due to polymerization (Egan et al., 1981).

It was also observed from Table (2) that the formaldehyde in cheese was not clearly changed during ripening period in each sample. Similar observation was obtained by Resmini et al. (1988) after 30 and 60 days of cheese ripened period by estimated spinacene. It can be revealed also that highly dangerous and toxic effect occurs because the cheese is consumed daily and long continuous intake of formalin might be harmful. Injection of formalin by humans has resulted in loss of consciousness, vascular collapse, pneumonia, haemorrhagic nephritis and abortion. formaldehyde cause damage to the gastrointestinal tract occurs primarily in the stomach and lower esophagus. The United States Environmental Protection Agency (EPA) has concluded that form aldehyde may be a carcinogen in humans (Haddad and Winchester, 1990). Formaldehyde is metabolized to formic acid and methanol; the formic acid may then be converted to methyl formate. It is also a metabolite of methanol. It combines rapidly with the amino-groups proteins and amino-acids (Clarke, 1978). severe acidosis results from rapid conversion of formaldehyde to formic acid (Matthew and Donald, 1988).

It can be recommended that formalin must not be added to the milk used in cheese manufcturing at any level and sufficient care should be taken at the farm to deliver it fresh to the cheese plants, also,

the milk of cheese must be pasteurized and active starter should be added before manufacture of cheese.

REFERENCES

Abdel-Mottaleb, L.; I.G. Abo-Elnaga and S.M. El-Gendy (1968): Studies on the effect of added formalin in the manufacture of Domiatti cheese. bull. Sci. Techn., 11: 207.

Atherton, H.V. and J.a. Newlander (1977): Chemistry and testing of Dairy products. (Fourth Edition). AVI Publishing Company, INC. Westport, connecticut, USA, p. 207.

Berg, J.C.T. Van cen (1988): Dairy technology in the tropics and subtropics. Centre for Agriculture Publishing and Documentation (Pudoc), Wageningen, the Netherlands.

Bottazzi, V.; Battisotti, b.; Chiusa, P. and Scolari, G.L. (1984): Synergistic action of formaldehyde and lysozyme for control of late blowin in Grana cheese. Scienza e Tecnica Lattiero-Casearia, 35 (4): 377-383. (Cited from Dairy Sci. Abs. 47: 3549).

Clarke, E.G.C. (1978): Isolation and Identification of Drugs. The Pharmaceutical Press, London, p.6.

Egan, H.; R.S. Kirk and R. Sawyer (1981): Pearson's chemical analysis of food. (Eighth Edition), churchill Livingstone, Edinburgh, London, Melbourne and N.Y., 1981, p. 78.79.

El-Gendy, Sh. M.; L. Abdel-Mottaleb and I. Abo-Elnaga (1980): Characteristics of Domiatti cheese made from milk containing formalin. Journal of Food Protection, 43: 694.

Haddad, L.M. and J.F. Winchester (1990): clinical Management of poisoning and drug overdose. W.B. Saunden Company, Philadelphia, PA 19106.