Chemical Composition and Occurrence of Mycotoxins in Six Blue-Mould-Ripened Cheeses in Egypt

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ABSTRACT

Chemical composition (fat, moisture, protein, salt, titratable acidity, and pH), proteolysis (free amino acids), lipolysis (total volatile fatty acids and free fatty acids), and some mycotoxins were determined in all cheese samples studied. Saint Agur was characterised by its high concentrations of total protein (22.04%), total amino acids (5.21mg/g), fat (33.79%), and TVFA (25.3 ml NaOH/100 mg cheese) and lowest concentrations of salt (2.41%) and moisture (40.20٪). Bleu d'Auvergne, had a pH around 6.0, and the highest concentrations of salt in moisture (8.0%), soluble nitrogen (51.98%), moisture (47.09%) and salt (3.76%) of all cheeses investigated. Bleu d'Auvergne had the lowest fat content (28.42%) and TVFA (20.1 ml NaOH/100mg cheese) of all the cheeses studied.

Cheese samples were analyzed for seven mycotoxins (Roqurfortine C, Mycophenolic acid, Penicillin acid, Aflatoxin G1, Aflatoxin M1, Aflatoxin B1, Aflatoxin B2). Roqurfortine C was detected in all of the blue mould cheese samples in concentrations of 1.6-14 μg kg^{-1}. Two blue-cheese samples (Saint Agur and Moby blue) contained also 0.35 and 0.47 μg kg^{-1} mycophenolic acid, respectively. Penicillic acid was detected (0.12, 0.41 and 0.53^{-1} μg/ kg) in Gorgonzola, Saint Agur and Moby blue, respectively. Aflatoxin G1, aflatoxin M1, aAflatoxin B1 and aflatoxin B2 were not detected in the tested cheeses, this is likely to be due to best manufacturing practises applied in the cheese production, and at the same time the storage conditions are all strictly controlled in order to avoid mould contamination and aflatoxins formation.

Keywords: blue cheese, chemical properties, ripening indices, aflatoxins, mycotoxins, organoleptic properties.

INTRODUCTION

Blue-type cheeses, characterised by the growth of *Penicillium roqueforti* in fissures throughout the cheese matrix, are one of the most easily identifiable cheese types. During ripening, blue-type cheeses undergo extensive proteolysis and lipolysis resulting in odour, flavour, appearance and texture development (Fox *et al*., 2000). The production of blue cheese with typical high quality flavour is mainly influenced by milk type, species of moulds used, rate of starter growth and salt content. It is well established that cheese ripening is a complex process involving the breakdown of the curd by proteolysis, lipolysis and other enzymes-catalyzed reactions to give the flavour and texture changes typical of the different cheese varieties. These reactions are brought about by enzymes present in the milk, added rennet, starter cultures, the moulds and yeasts and the cheese milk micro flora (Kinsella & Hawng 1976). Lactic acids bacteria were reported to the dominant group from salting on words, while moulds and yeasts were dominant during ripening (Lopez-Diaz *et al*., 1996). *Penicillium roqueforti* has been used as a secondary starter culture for the ripening of blue–mould cheeses, such as Gorgonzola, Danabule, Roquefort, Bleu de Bresse and Stilton for centuries. Because of the coevolution with lactic acid bacteria, *Penicillium roqueforti* is resistant to lactic acid, acetic acid, carbon dioxide and several other lactic acid bacterial metabolites (Boysen *et al*., 2000).

There are many false reports on mycotoxin production from *Penicillium roqueforti*. Mycotoxins are toxic secondary metabolites produced by many species of fungi. The presence of these substances in dairy products can have two origins; (a) indirect contamination, which results from lactating animals ingesting contaminated feed and (b) direct contamination, which occurs because of intentional or accidental growth of moulds on dairy products (van Egmond 1989). Some of the fungal starters used in the cheese manufacture have been shown to produce various mycotoxins on culture media. *P. roqueforti*, widely used as a starter in the blue mould cheeses, is known to synthesise Roquefortine C, PR-toxin, mycophenolic acid and isofumigaclavins (Samson & Frisvad, 2004). The occurrence of Roquefortine C and PR toxin in domestic and imported blue cheeses in Italy, the identification of the penicillia used as starter, and the investigation of their capacity for producing toxins in culture media were investigated (Frisvad *et al*., 2004). Roquefortine C was always found in the cheeses with levels ranging from 0.05 to 1.47 µg kg^{-1}, whereas PR-toxin or penicillic acid could not find in blue cheeses (Schoch *et al*., 1984). Fungal strains present in the domestic cheeses included *Penicillium glabrum*, *P. roqueforti* and *P. cyclopium* in the Gorgonzola "dolce" and *P. roqueforti* in the Gorgonzola "naturale". (Erdogan *et al*., 2003, Samson & Frisvad, 2004). Other mycrobiota besides starters may be present in mould cheeses as contaminants, since various fungi are encountered in cheese factory environments and some of these organisms can be potential mycotoxin producers. Few reports indicating the occurrence of mycotoxins due to contaminating fungi in mould cheeses. Lafont *et al*., (1990) detected penicillic acid in one blue cheese sample of dubious organoleptic properties, indicating this might be because of contaminating fungi rather than actual starters.

The objectives of the present study were to determine the gross compositional constituents of a number of blue–type cheeses in Egyptian markets and to evaluate their nutritional values. The second goal of this study was to determine the level of mycotoxins in blue cheeses samples collected in order to evaluate its safety.

MATERIALS AND METHODS

Collection of samples

Six blue-type cheeses selected to represent a rang of different blue-moulds cheeses (2-4 months old), were purchased from the Egyptian supermarkets at Cairo and Alexandria. Cheeses included the following: Danish Blue (5 samples), Blue d'Auvergne (7 samples), Blue Stilton (4 samples), Gorgonzola (4 samples), Moby blue (6 samples) and Saint Agur (6 samples).

Chemical analysis

Gross chemical composition was analysed using standard methods; moisture contents were estimated by the oven drying method (AOAC, 1990). Salt content was determined according to (AOAC, 1990). Acidity content expressed as lactic acid was determined according to the procedure of (BSI) (1985). The modified Gerber method was used to determine fat in cheese samples as described by BSI (1985).The level of pH (pH 4.6-SN) was measured using the method of Kuchroo & Fox (1982). Free amino acids concentrations were determined using the method of Fenelon *et al*., (2000). The concentrations of free fatty acids (C2:0, C3:0, C4:0, C6:0) were determined using the method described by Kilcawley *et al*. (2001). Total volatile fatty acids of cheese samples were determined according to the method of Kosikowki (1978). Cheese samples were analysed in duplicate, and they are stored at 4°C prior to analysis.

Determination of mycotoxins

Samples preparation

The samples were prepared with a slight modification as described by the method of Rundberget & Wilkins (2002). Ten grams of cheese were weighed and 60 ml of acetonitrile (containing 0.1% formic acid) and 50 ml of hexane were added. The sample was homogenized by Ultra Turrax homogenizer for five minutes and centrifuged (4000 rpm, for 10 min,). The sample was filtered through an S & S 2329 filter paper and a 10ml portion of the acetonitrile phase was evaporated to dryness under a stream of nitrogen at +50°C. The residue was dissolved in 0.2ml of methanol and filtered through a 0.2 mm syringe filter into an auto-sampler vial.

Capillary liquid chromatography-tandem mass spectrometric (LC-MS/MS) analysis

Chromatographies were carried out on a Waters 2695 separation module equipped with a Waters 996 photodiode array detector (Waters, Millford, MA, USA), by injecting 10 µl of the sample on a Symmetry C18 column (2.1x100 mm, 3.5 µm; Waters, Milford, MA, USA). Elution was performed by using a gradient (74.9% water, 25% acetonitrile and 0.1% acetic acid, v/v/v), which were held for 16 min, then changed to (9.9% water, 90% acetonitrile and 0.1% acetic acid, v/v/v) for 24 min with flow rate 0.2 ml /min. LC was coupled to a Micro-Mass Quattro Micro triple-quadrupole mass spectrometer (Micro mass Ltd, Manchester, UK) equipped with an electrospray ionization (ESI) probe operating in the positive mode. Multiple reactions monitoring (MRM) mode was used for identification and quantification of the compounds. The retention times of the standard mycotoxins were 3.5, 4.2, 4.7, 8.2, 8.6, 11.3 and 27 min for Penicillic acid, Roquefortine

C, Aflatoxin M1, Aflatoxin B2, Aflatoxin G1, Aflatoxin B1 and Mycophenolic acid, respectively.

RESULTS AND DISCUSSION

Chemical composition

The chemical composition and the concentrations of free amino acids (FAA), free fatty acids (FFA), in six blue-mould cheeses are shown in Tables (1, 2 and 3), respectively. Saint Agur was characterised by its high concentrations of total protein, total amino acids and free amino acids. Of the cheeses samples analysed, Saint Agur had the highest concentrations of total protein (22.04%), total amino acids (5.211 μg g^{-1}) and most individual free amino acids (Tables 1 and 2). Saint Agur had a relatively high level of soluble nitrogen (48.47%) indicating a high rate of overall proteolysis. Considerable proteolysis occurs in blue-type cheese (Hewedi & Fox, 1984) resulting from low cook temperatures and a high rate of acid production during the manufacture, and the action of rennet, proteinases from secondary microflora such as *P. roqueforti*.

Saint Agur and Blue Stilton had relatively low concentrations of moisture (40.20 and 41.08%, respectively), salt (2.41 and 2.58%, respectively) and salt in moisture (6.0 and 6.28%, respectively), relatively high fat content (33.79 and 32.50%, respectively), total protein (22.04 and 21.80%, respectively), soluble nitrogen value (48.47 and 46.82%, respectively) and high concentrations of fat in the dry matter (56.50 and 55.15%, respectively) (Table 1).Bleu d'Auvergne a French blue-type manufactured using cow's milk, had a pH of 5.9, and the highest concentrations of salt (3.76%), salt in moisture (8.00%), soluble nitrogen (51.98%), moisture (47.09%). Bleu d'Auvergne had the lowest fat content (28.42%), TVFA (20.1 ml NaOH 0.1N /100μg) and total protein (19.70%) of all the cheeses studied (Table 1). Gorgonzola, an Italian blue cheese had a lowest concentration of protein (19.43%), protein in the dry matter (35.00%) soluble nitrogen (44.32%) and TVFA (22.21 ml NaOH 0.1N /100μg cheese).

The extensive proteolysis that occurs in blue-type cheeses is caused, primarily, by the proteolytic system of the secondary microflora (*P. roqueforti*) (McSweeney & Sousa, 2000). Factors affecting the degree of proteolysis include the mould strain and the ripening time (Larsen, *et al*., 1998).

Data given in Table (2) indicate that glutamic acid is the most predominant free amino acid in the six blue-type cheese under study. However, considerable differences in glutamic acid content could be traced. Saint Agur cheese exhibited the highest glutamic acid content (9280 μg g^{-1}), while, Gorgonzola cheese possessed the lowest content of glutamic acid being 3713 μg g^{-1} cheese.

Table 1: Gross compositional analysis of blue-type cheeses

Chemical properties	Bleu d'Auvergne	Saint Agur	Blue Stilton	Gorgonzola	Danish blue	Moby blue
Moisture*	47.09±1.6	40.2±1.1	41.08±1.0	44.47±1.5	41.58±1.3	42.42±1.2
Protein*	19.7±0.35	22.04±0.5	21.80±0.43	19.43±0.37	21.6±0.39	21.50±0.41
Fat*	28.42±0.9	33.79±0.5	32.50±0.73	31.80±0.7	31.4±0.6	32.4±0.5
Salt*	3.76±0.2	2.41±0.22	2.58±0.12	3.0±0.16	3.05±0.16	2.75±0.15
pH	5. 90	5.59	5.64	5.37	5.28	5.87
Titratable acidity**	2.01±0.15	2.15±0.18	2.15±0.25	2.25±0.22	2.35±0.19	2.00±0.2
P/DM	37.23	36.86	37.05	35.00	37.62	37.34
F/DM	53.73	56.50	55.15	57.27	54.68	55.57
S/M	8.00	6.00	6.28	6.75	7.16	6.48
TVFA	20.1±0.57	25.3±0.50	23.9±0.3	22.2±0.39	23.2±0.29	23.4±0.4
SN	51.98±1.9	48.47±1.5	46.82±1.8	44.32±1.4	48.08±1.2	47.00±1.1

* Values expressed as percentage. ** Values expressed as % of lactic acid.
P/DM = protein in the dry matter. F/DM=fat in the dry matter.
S/M = salt in moisture SN= water soluble nitrogen.
TVFA = total volatile fatty acids expressed as (ml 0.1 NaOH/100mg)

Table 2: Free amino acid analysis of six blue-type cheeses

Free amino acids	Bleu d'Auvergne	Saint Agur	Blue Stilton	Gorgonzola	Danish blue	Moby blue
Ala	857	2723	1778	645	1144	1854
Asp	1534	1936	1207	701	1290	1379
Cys	488	481	523	389	490	459
Glu	4569	9280	7226	3713	6415	5735
Gly	457	881	712	379	603	583
His	1626	2151	1713	740	2269	1673
Ile	1600	2635	2000	947	1367	1765
Leu	3689	6037	5248	3345	4748	4944
Lys	4376	7112	5313	2821	4803	4677
Met	1062	1957	1604	1000	1679	1321
Phe	2000	3226	2600	1525	2109	2415
Pro	1146	4100	2359	843	1839	2009
Ser	2115	2635	1891	1915	3568	2231
Thr	877	1550	1240	523	926	983
Tyr	1897	1313	1760	1492	1616	1649
Val	2317	4060	3235	1902	2345	3230
Total	30664	52108	41215	22857	37219	36905

Expressed as $\mu g\ g^{-1}$ cheese

Table 3: Free fatty acid content in different blue-type cheeses

Free fatty acids	Bleu d'Auvergne	Saint Agur	Blue Stilton	Gorgonzola	Danish blue	Moby blue
Butyric acid	1565	12204	6901	1244	5151	4579
Propionic acid	87745	51	101	ND	ND	77
Hexanoic acid	10916	7902	3141	332	2505	2945
Acetic acid	801	1817	1721	109	1501	974

Expressed as $\mu g\ g^{-1}$ cheese ND = not detected

Lysine was found to be the second predominant free amino acid in four types out of the six ones investigated in the present study. Notwiththstanding, the second predominant free amino acid in the other two types (Gorgonzola and Moby Blue) is leucine being 3345 and 4944 $\mu g\ g^{-1}$ cheese, respectively (Table 2).

It was obvious that leucine is the third predominant free amino acid in four out of the six types of cheese under study. It varied from 3689 $\mu g\ g^{-1}$ cheese for Bleu d'Auvergne to 6037 $\mu g\ g^{-1}$ for Saint Agur cheese (Table 2).

It is worth to mention that the six types of cheese under study varied considerably in terms of the least free amin acid content therein. It is glycine in Bleu d'Auvergne (457 $\mu g\ g^{-1}$), Gorgonzoia (379 $\mu g\ g^{-1}$) and Danish blue cheese (603 $\mu g\ g^{-1}$). On the other hand, cysteine was found to be the least free amino acid in Saint Agur cheese (481 $\mu g\ g^{-1}$), Bleu Sitlton cheese (523 $\mu g\ g^{-1}$) and Moby blue cheese (459 $\mu g\ g^{-1}$) as its shown in Table (2).

The six types of cheese under study can be ascendingly arranged regarding the total content of free amino acids as follows: Saint Agur (52108 $\mu g\ g^{-1}$), Blue Stilton (41215 $\mu g\ g^{-1}$), Danish Blue (37219 $\mu g\ g^{-1}$), Moby Blue (36905 $\mu g\ g^{-1}$), Bleu d'Auvergne (30664 $\mu g\ g^{-1}$) and finally Gorgonzola cheese (22857 $\mu g\ g^{-1}$) (Table 2).

Data presented in Table (3) reveal that butyric acid is the most predominant free fatty acid present in five out of six types of cheese investigated in the present study. It varied from 1244 µg g^{-1} of Gorgonzola cheese to 12204 µg g^{-1} of Saint Agur cheese. However, propionic acid was the most predominant free fatty acid in Bleu d'Auvergne cheese being 87745 µg g^{-1}. In contrast, propionic acid could not be detected in any of Gorgonzola or Danish Blue cheeses. Meanwhile, hexanoic acid varied considerably in a range from 332 µg g^{-1} of Bleu d'Auvergne cheese to 10916 µg g^{-1} of Blue d'Auvergne cheese. Noticeable differences could be pointed out regarding the acetic acid content in the investigated six types of cheese. It varied from 109 µg g^{-1} of Gorgonzola cheese to 1817 µg g^{-1} of Saint Agur cheese (Table 3).

Mycotoxins in cheese samples

The mycotoxins concentration of the cheeses studies are presented in Table (4). All the blue cheeses contained Roquefortine C from 1.6 to 14µg g^{-1}. Mycophenolic acid was also found 0.35 and 0.47µg kg^{-1} in Saint Agur and Moby blue respectively. Penicillic acid was detected (0.12, 0.41 and 0.53µg kg^{-1} respectively) in Gorgonzola, Saint Agur and Moby blue. Other investigated mycotoxins were not detected in the samples. The Roquefortine C (1.6–14µg kg^{-1}) and mycophenolic acid (0.3-0.47µg kg^{-1}) concentrations found in the samples were mostly in accordance with the previous studies (Lafont *et al*., 1990, Bentley, 2000, Finoli, *et al*., 2001,

Erdogan *et al*., (2003), Samson & Frisvad (2004). Schoch *et al*. (1984) reported Roquefortine C at levels from 0.2–2.29µg kg^{-1} and Finoli *et al*. (2001) levels from 0.05–1.47µg kg^{-1} in various blue cheeses. Compared to these results, the concentration of 14 µg kg^{-1} Roquefortine C in Blue d'Auvergne cheeses is quite high. In the study of Lafont *et al*. (1990), the estimated concentrations of mycophenolic acid were between 0.01 and 15.0 µg kg^{-1} in blue cheeses. Both blue cheese samples Saint Agur and Moby blue contained a detectable amount of mycophenolic acid; it is evident that this toxin can be avoided with the selection of the starter strain, as proposed also by Lafont *et al*. (1990). The other investigated mycotoxins were absent in the cheese samples collected. This is likely to be due to best manufacturing practises applied in the cheese production. The raw materials, the manufacturing processes and the storage conditions are all strictly controlled in order to avoid mould contamination and mycotoxin formation (Engel & Tauber, 1989).

REFERENCES

AOAC, **1990**. Official Methods of Analysis (15th Ed). Association of Official Analytical Chemists. Washington, DC.

Bentley, R. **2000**. Mycophenolic acid: A one hundred year Odyssey from antibiotic to immunosuppressant. Chemical Reviews **100**: 3801–3825.

Boysen, M., Jacobsson, K.G. & Schnuer, J. **2000**. Molecular identification of species from the *P. roqueforti* group associated with spoiled animal feed. Appl. Environment Microbiology, **66**: 1523-1526.

BSI **1985**. British Standard Method Institute for chemical analysis of cheese, Gerber method for the determination of fat in milk and milk products. B.S. 696 parts I and II. London: British Standards Institute.

Engel, G. & Teuber, M. **1989**. Toxic metabolites from fungal cheese starter cultures (*Penicillium camemberti* and Penicillium roquef orti). In: Mycotoxins in Dairy Products. van Egmond HP, (Ed.) London: Elsevier applied science. pp 163–192.

Table 4: Amounts of detectable mycotoxins in the examined blue-type cheeses

Designation	Country of origin	Roque-fortine C	Mycophe-nolic acid	Penicillic	Aflatoxin G_1	Aflatoxin M_1	Aflatoxin B_1	Aflatoxin B_2
Blue d'Auvergne	France	14	ND	ND	ND	ND	ND	ND
Danish Blue	Denmark	6.5	ND	ND	ND	ND	ND	ND
Blue Stilton	England	4.6	ND	ND	ND	ND	ND	ND
Gorgonzola	Italy	1.6	ND	0.12	ND	ND	ND	ND
Saint Agur	France	2.7	0.35	0,41	ND	ND	ND	ND
Moby blue	Egypt	9.4	0.47	0.53	ND	ND	ND	ND

ND = not detected.

Erdogan A., Gurses M. & Sert S. **2003**. Isolation of moulds capable of producing mycotoxins from blue mouldy Tulum cheeses produced in Turkey. International Journal of Food Microbiology, **85**: 83–85.

Finoli, C., Vecchio, A., Galli, A. & Dragoni, I. **2001**. Roquefortine C occurrence in blue cheese. Journal of Food Protection. **64**: 246-251.

Fox, P.F., Guinee, T.P., Cogan, T.M. & McSweeney, P.L.H. **2000**. Fundamentals of Cheese Science. Gaithersburg, Maryland, USA; Aspen Publishers, Inc.

Frisvad, J.C., Smedsgaard, J., Larsen, T.O. & Samson, R.A. **2004**. Mycotoxins, drugs and other extrolites produced by species in P. subgenus and P. Strud. Mycol. **49**: 201-241.

Gripon, J.C. **1993.** Mould- ripened cheeses. In: Cheese Chemistry, Physics and Microbiology. II Major Cheese Groups. Fox, P.F. (ed), Elsevier Science Publisher Ltd, Cambridge, England.

Hewedi, M.M, & Fox, P.F. **1984**. Ripening of blue cheese: Characterization of proteolysis. Milchwissenschaft, **39** (4): 198–201.

Kilcawley, K.N., Wilkinson, M.G., & Fox, P.F. **2001**. A survey of lipolytic and glycolytic end-products in commercial Cheddar enzyme-modified cheese. J. of Dairy Sci., **84** (1): 66–73.

Kinsella, J.E. and Hwang, D. **1976**. Biosynthesis of flavours by *P. roqueforti*. Biotechnology Bioengeneering, **18**: 927-938 .

Kosikowski, F.V. **1978**. Cheese and Fermented Milk Foods. II (2nd ed.) Edwards Brothers Inc. Ann. Arbor, MI.

Kuchroo, C.N. & Fox, P.F. **1982**. Soluble nitrogen in cheese: Comparison of extraction procedures. Milchwissenschaft, **37**(6): 331–335.

Lafont, P., Siriwardana, M.G. & DeBoer, E. **1990**. Contamination of dairy products by fungal metabolites. Journal of Environmental Pathology, Toxicology and Oncology., **10** (3): 99-102.

Larsen, M.D., Kristiansen, K.R. & Hansen, T.K. **1998**. Characterization of the proteolytic activity of starter cultures of *P. roqueforti* for production of blue veined cheeses. International Journal of Food Microbiology, **43** (3): 215–221.

Lo´pez-Dı´az,T.M., Roma´n-Blanco, C., Garcı´a-Arias, M.T., Garcı´a-Fernandez, M.C. & Garcı´a-Lo´pez, M.L. **1996**. Mycotoxins in two Spanish cheese varieties. International Journal of Food Microbiology, **30**: 391–395.

McSweeney, P.L.H. & Sousa, M.J. **2000**. Biochemical pathways for the production of flavour compounds in cheese during ripening: A review. Le Lait, **80** (3): 293–324.

20-Rundberget, T. & Wilkins A.L. **2002**. Determination of Penicillium mycotoxins in foods and feeds using liquid chromatography–mass spectrometry. J. of Chromatography A, **964**: 189–197.

Samson, R.A. & Frisvad, J.C. (Edts) **2004**. Penicillium subgenus penicillium: New taxonomic schemes and mycotoxins and other extrolites (Utrecht: Centraalbureau voor Schimmel cultures).

Schoch U., Lu¨thy J., & Schlatter C. **1984**. Mykotoxine von P.roqueforti and P. camemberti in Ka¨se. I. Vorkommen chemisch identifizierter Mykotoxine. Milchwissenschaft **39** (2): 76–80.

van Egmond, H.P. (Ed.) **1989**. Introduction. In: Mycotoxins in Dairy Products. London: Elsevier Applied Science. pp1-9.

التركيب الكيماوي وتواجد السموم الفطرية في ستة أنواع من الجبن المسوى بالفطريات الزرقاء في مصر

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درس التركيب الكيماوي (الدهن، الرطوبة، البروتين، الملح، الحموضة التنقيطية، رقم الأس الهيدروجيني pH) وكذلك تحلل البروتينات (الأحماض الأمينية الحرة) وتحلل الدهون (الأحماض الدهنية المتطايرة الكلية، الأحماض الدهنية الحرة)، كما تم تقدير بعض السموم الفطرية في عينات الجبن موضع الدراسة.

تبين أن نوع الجبن Saint Agur يتميز بمحتواه العالي من البروتين (٢٢,٠٤٪)، الأحماض الأمينية ٥,٢١ ملجم/جم، الدهن (٣٣,٧٩٪) الأحماض الدهنية المتطايرة الكلية (٢٥,٣ مل NaOH/ ١٠٠ ملجم جبن) بينما يحتوي على أقل تركيز من الملح (٢,٤١٪) والرطوبة (٤٠,٢٪). أما نوع الجبن Bleu d'Auvergne فكان رقم الأس الهيدروجيني له حول الرقم ٦، واحتوى على أعلى تركيز ملح في الرطوبة (٨,٠٪) والنيتروجين الذائب (٥١,٩٨٪)، الرطوبة (٤٧,٠٩٪)، الملح (٣,٧٦٪) وذلك مقارنة بالأنواع الأخرى موضع الدراسة، كما احتوى هذا النوع من الجبن على أقل محتوى دهن (٢٨,٤٢٪) وأحماض دهنية متطايرة كلية (٢٠,١ مل NaOH/ ملجم جبن) مقارنة بالأنواع الأخرى من الجبن التي تم اختبارها وقد تم تقدير سبعة من السموم الفطرية في أنواع الجبن الستة، واشتملت هذه السموم على ما يأتي : Aftlatoxin B2 ، Aflatoxin B1 ، Aflatoxin M1 ، Aflatoxin G1 ، Penicillin acid ، Mycophenolic acid ، Requrfortine C. وقد تم الكشف على سم الـ Roqurfortine C في كل أنواع الجبن المختَرة وذلك بتركيزات تراوحت بين ١,٦، ١٤,٠ ميكروجرام/كجم. كذلك فقد احتوى نوعان من الجبن وهما الـ Saint Agur والـ Moby blue على السم الفطري Mycophenolic acid وذلك بتركيزي ٠,٣٥، ٠,٤٧ ميكروجرام/كجم على الترتيب. من ناحية أخرى فقد تم الكشف عن هذا السم بتركيزات ٠,١٢، ٠,٤١، ٠,٥٣ ميكروجرام وذلك في أنواع ثلاثة من الجبن وهي : Gorgonzola ، Saint Agur ، Moby blue على الترتيب. وأوضحت الدراسة عدم احتواء أي نوع من أنواع الجبن المختبرة على أي من سموم الأفلاتوكسينات الأربعة، وهو ما يؤكد أن عملية تصنيع هذه الأجبان وكذا تخزينها كانت جيدة وحالت دون حدوث أي تلوث بالفطريات وتكوين سموم الأفلاتوكسينات.