

Deleterious effect of high ammonia percentage in ration on productivity of sheep and goats

Arafa, M. M. * Hanan, A. Azoz, Riham, M. Rafat and Hanaa, A. Allam**

* Biochemistry Dept., Animal Health Research Institute –Dokki-

**udder and neonatal disease Research dept ,animal reproduction research institute - El Haram.

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Abstract

A private farm of adult sheep and goats in El Menofia governorate showed drop in milk production and rapid emaciation associated with ammonia like odour in the farm. Analysis of soil, bedding straw, ration and hay indicated presence of high percentage of ammonia. Complete blood picture, serum ammonia, urea, glucose, AST, ALT, T-protein, albumin, globulin, creatinin, uric acid, calcium, magnesium and phosphorous concentrations were determined. Milk fat, protein, lactose, milk urea, nitrogen, total solids and solids not fat% and somatic cell count were determined. The obtained results showed presence of high levels of ammonia in blood with significant changes in most of estimated serum parameters. However, the blood picture displayed presence of anemia and leucocytopenia in the affected animals compared with the clinically normal animals. Milk results recorded significant decreased levels of fat, protein and total solids, while milk urea nitrogen increased significantly in both sheep and goats groups.

Key words: ammonia, sheep, goat, milk, serum.

Introduction

Air pollution is considered to be one of the most important problems in the animal production especially in housing under intensive system. It consists of the gaseous products of animal respiration and microbiological decomposition products of manure together with air borne dust and associated disease agents derived from bedding, food and from animals themselves. Ammonia toxicity is the same in all mammals, and all species of domestic animals are susceptible to ammonia toxicity **Yousif (1984)** and **Roller et al., (1982)**. The individual role of ammonia in the development of respiratory disease remains unclear since it acts synergistically with other pollutants and may influence the incidence and severity of biological agent inducing respiratory diseases. Ammonia is highly soluble in water and is

presumably largely absorbed by the distal airway mucus.

Ammonia can favor bacterial contamination of the lungs by decreasing pulmonary clearance and inducing airway mucosal inflammation, as reported by **Drummond et al., (1981)**.

In normal healthy population, the level of blood ammonia ranged from 48 to 90 ug/dl, but with workers in nitrate fertilizer factories it ranged from 124 to 270 ug/dl with no signs of toxicity as stated by **seleem et al., (1985)**. A chance of toxicity occurring was extremely high when the blood ammonia concentration exceeded above 0.8 mg/dl (**Bartley et al., 1976**). It has been suggested by **Asmar et al., (2001)** that ammonia exposure increases inflammatory, immune and neuroendocrine stress responses in pigs. The deleterious effects of ammonia on weight gain, feed intake and

feed conversion of broiler chickens have been reviewed by **Carlile (1984)**, **Whyte (1993)** and **Kirstensen and Wathes (2000)**. Atmospheric ammonia is believed to cause release of cytokines by alveolar macrophages and neutrophils, constituting a potent inflammatory response as mentioned by **Murata and Horino (1999)**. The clinical signs of ammonia toxicity in sheep, goats and cattle are uneasiness, dullness, muscle and skin tremors, excessive salivation, frequent urination and defecation, rapid respiration, incoordination, stiffening of the front legs, prostration, tetany and death as reported by **Helmer and Bartely (1971)**. The clinical signs of ammonia exposure in sheep are drop in production and rapid emaciation reported by **Kubesy (1987)**. The ammonia exists in two forms, free ammonia (NH_3) at higher PH and as ammonium ion (NH_4) at lower PH. **Visek (1968)** observed that tissue membranes are permeable to the lipid soluble NH_3 form but impermeable to the charged NH_4 ions, so absorption of ammonia will be greater at higher PH than at lower PH. The present study is a trial to clarify the effects of ammonia toxicity on some biochemical and hematological parameters, thyroid hormones

and glucose level and milk composition in dairy sheep and goats.

Materials and Methods

Twenty-five dairy sheep and goats (Baladi) (50 animals) from a private farm at El Menofia governorate, showed clinical symptoms of severe emaciation and drop in milk production with ammonia like odour in the farm. All animals were fed a concentrate ration and hay twice daily at the same time of milking. Five clinically healthy sheep and goats (baladi) from another farm were fed a concentrate ration and hay twice daily were used as control. Samples from soil, bedding straw, ration and hay were analyzed for estimation of ammonia according to **Mondzac *et al.*, (1965)**. Two blood samples were collected at the same time from each animal (100 samples) from jugular vein. The first blood samples were collected in heparinized test tubes to be used for determination of complete blood picture (**RBC, Hb, PCV and WBCs**) according to **Coles (1986)**. The second blood samples were collected into dry and clean test tubes and left to separate serum samples which were used for estimation of the following parameters (Table 1).

Parameter	According to the method described by
Ammonia	Mondzac <i>et al.</i> (1965).
Urea	Patton and Cruch (1977).
Creatinine	Seeling and Wust (1969).
T. Protein	Sonnenwirth and Jarett (1980).
Albumin	Drupt (1974).
Uric Acid	Fossati, <i>et al.</i> (1980).
ALT	Reitman and Frankel (1957).
AST	Reitman and Frankel (1957).
Calcium	Glindler and King (1972).
Phosphorus	Killichling and Freiburg (1951).
Magnesium.	Neilly and Nelly (1956).
Glucose	Werner <i>et al.</i> (1970).

Milk Samples.

Fifty milk samples were collected from the affected farm (25 sheep and 25 goats), and 10 milk samples were collected from clinically healthy animals (5 sheep and 5 goats) from another farm. Each sample was collected in a clean, sterile and dry McCartney glass and preserved in ice tank. The samples were analyzed for somatic cell count (S.C.C.) and milk parameters (fat %, protein%, lactose%, milk urea nitrogen%, total solids% and solids not fat%).

Field Test (California Mastitis Test, CMT).

That test was applied According to **APHA (1992)** for detection of subclinical mastitis in dairy ewes, California mastitis test was performed on individual milk samples collected from each half udder of every dairy ewes to detect subclinical mastitis. The C.M.T gives an indirect estimation of S.C.C and it is based upon a gelling reaction between the nucleic acid of the cells and a detergent reagent depending on the amount of gel formation.

Measurements of milk parameters.

It was done by using infra milk analyzer 150, from Bentley instruments.

Milk urea nitrogen (MUN).

It was calculated by multiplying milk urea concentrations by 0.467, according to **Batton and Crouch (1979)**.

Somatic cell count.

It was estimated automatically using SOMA-COUNT 150, from Bentley.

Statistical analysis.

The obtained data were statistically analyzed using T student test according to **SPSS 14 (2006)**.

Results**Hematological and biochemical parameters:**

All hematological parameters (Hb, RBCs, PCV% and WBCs) of the affected sheep and goats decreased significantly comparing with control ones (Table2).

Results of serum biochemical parameters (Table3) exhibited significant increased levels of ammonia, albumin, uric acid, AST, ALT activities and glucose in both sheep and goats except urea concentration recorded high significant level in sheep only comparing with control animals. Creatinine and total protein concentrations did not show any significant difference between different groups. Serum globulin concentration decreases significantly and consequently A/G increased significantly in affected animals.

Levels of serum calcium, magnesium and phosphorus recorded significant lower concentrations in all tested animals compared with control ones.

Milk parameters.

Result of California mastitis test (CMT) exhibited negative results in all examined samples. Our results of milk parameters (Table4), revealed significant elevated levels of milk urea nitrogen (MUN) (sheep and goats) and somatic cell count (goats only), meanwhile fat, protein and total solids percentage decreased significantly in both sheep and goats comparing with healthy animals. Solids not fat levels did not show any significant change.

Results of ammonia analysis in soil, bedding straw, ration and hay revealed high percentages of ammonia which were 105%, 43%, 10% and 13 %, respectively (Table5).

Table (2). Hematological picture of control and affected sheep and goats.

Animals	Sheep		Goats	
Parameters	Healthy (control)	Affected	Healthy (control)	Affected
Hb (gm/dl)	12.19 ± 0.11	5.90 ± 0.40 **	13.7±0.23	6.2±0.92**
RBCs×10 ⁶ /cum	7.67 ± 0.11	4.73 ± 0.14 **	6.8±0.49	4.9±0.65**
PCV %	31.00 ± 0.65	21.71 ±0.92**	34.2±1.1	20±0.94**
WBCs×10 ³ /cum	8.54 ± 0.15	4.19 ± 0.09 **	9.33±0.39	6.2±0.11 **

**Significant at p<0.01

Table (3). Serum biochemical profile of affected and clinically healthy sheep and goats.

Animals	Sheep		Goats	
Parameters	Healthy (control)	Affected	Healthy (control)	Affected
Ammonia (mg/dl)	0.13 ± 0.01	0.9 ± 0.04 **	0.13±0.01	0.86±0.05**
Urea (mg/dl)	22.29±0.97	25.57 ± 1.13*	23±1.6	24.6±1.8
Creatinine (mg/dl)	1.16 ± 0.05	1.219±.014	1.2±0.07	1.25±0.08
Albumin (g/dl)	2.49±0.14	3 ± 0.14 *	2.5±0.22	3.2±0.11 *
T. Protein (g/dl)	6.5 ± 0.13	6.16± 0.15	6.4±0.31	6.2±0.22
Globulin (g/dl)	4.01± 0.23	3.16± 0.14**	3.9±0.07	3±0.05**
A/G ratio	.621 ± 0.06	.949±0.41 **	0.641±.07	1.666±.09**
Uric Acid (mg/dl)	2.21 ±0.07	10.83± 2.02 **	2.3±0.33	6.9±0.21**
AST (IU/ml)	21.68 ± 1.72	99± 3.22 **	22±0.74	80±0.95**
ALT (IU/ml)	10.00 ± 0.9	79±0.34 **	13±2.1	67±1.33**
Calcium (mg/dl)	10.46 ± 0.12	6.91±0.19**	10.3±0.82	7.2±0.67*
Inorganic Phosphorus (mg/dl)	6.37 ± 0.28	2.3± 0.11 **	6.5±1.1	2.4±0.29**
Magnesium (mg/dl)	3.2 ± 0.09	1.08± 0.04 **	3.1±0.94	1.02±0.17**
Glucose (mg/dl)	64.14 ± 1.88	143.57± 6.58 **	70±1.9	140±1.21**

* Significant at P<0.05

** Significant at P< 0.01

Table (4). Milk parameters of affected and clinically healthy sheep and goats.

Animals	Sheep		Goats	
Parameters	control	Affected	control	Affected
Fat % Mean $\pm$ S.E.	5.43 $\pm$ 0.63	***2.43 $\pm$ 0.1	4.2 $\pm$ 0.08	***2.59 $\pm$ 0.14
Range	(4.19-5.2)	(1.12-3.55)	(3.9 – 4.4)	(1.47- 4.32)
Protein % Mean $\pm$ S.E.	5.418 $\pm$ 0.32	***3.75 $\pm$ 0.21	3.32 $\pm$ 0.085	***2.44 $\pm$ 0.044
Range	(4.7-6.14)	(2.46-5.34)	(3.1 – 3.6)	(2.01-2.89)
Lactose % Mean $\pm$ S.E.	3.46 $\pm$ 0.15	3.11 $\pm$ 0.43	4.39 $\pm$ 0.051	4.41 $\pm$ 0.027
Range	(2.15-4.16)	(2.07-4.16)	(4.21 – 4.48)	(4.21-4.74)
MUN mg % Mean $\pm$ S.E.	12.11 $\pm$ 0.43	***14.4 $\pm$ 0.3	9.00 $\pm$ 0.35	***12.79 $\pm$ 0.625
Range	(9.5-12.74)	(12.37-16.57)	(9.52-9.89)	(9.52-15.9)
Total Solids (T.S) % Mean $\pm$ S.E.	11.3 $\pm$ 0.21	***9.97 $\pm$ 0.24	12.18 $\pm$ 0.102	***10.34 $\pm$ 0.175
Range	(10.52-11.8)	(8.51-11.75)	(11.85-12.5)	(8.84-12.53)
Solids Not Fat (S.N.F.) % Mean $\pm$ S.E.	6.86 $\pm$ 0.28	7.53 $\pm$ 0.16	7.96 $\pm$ 0.061	7.75 $\pm$ 0.053
Range	(6.2-7.8)	(6.1-8.71)	(7.76-8.14)	(7.22-8.21)
SCC X 10 ³ Mean $\pm$ S.E.	71 $\pm$ 16	77 $\pm$ 9	77 $\pm$ 16	**140 $\pm$ 8
Range	(42-111)	(40-199)	(22-110)	(80-202)

**significant at p<0.01

***significant at p<0.001

Table (5). Percentage of ammonia in soil, bedding straw, ration and hay.

	Ammonia %	Permissible limit of ammonia according to A.O.A.C (1990)
Soil	105%	Up to 10%
Bedding straw	43%	Up to 5%
Ration	10%	Up to 5%
Hay	13%	Up to 5%

Discussion

The significant decrease in PCV%, RBCs and Hb levels reported in (Table 1) are on line with observation of **Fafioye (2002)** who noted a significant decrease in the blood cell parameters due to stress of exposure to ammonia. The decreased value in PCV may be due to anemia and haemodilution or haemolysis of RBCs. as reported by **Fafioye (2002)**. also **Saeed (1997)** recorded a marked reduction in RBCs count and Hb concentration.

The significant decrease was observed in white blood cell count may be due to the stress of exposure to sublethal level of ammonia as recorded by **Maule and Schreck (1990)** who noted that stress affects the number of leukocytes in immune organs.

Our results recorded an increase in serum glucose level which was concided with results of **Bakry (1999)** and **Mehra et al., (2005)**. This increase may be due to mobilization of energy from all available sources to combat the stress as reported by **Colombo et al., (1990)**. **Donald, et al., (1981)** suggested that hyperglycemic effect of ammonia appears to be an indirect result of an increased output of hormones. This explanation was supported by **Wiecheteck, et al., (1988)** who mentioned that the hyperglycemic effect completely depends on the neurogenic increase of adrenal medullary hormones, in general short induced stress increases metabolic rate with increase in glucose and urea levels.

Serum biochemical results showed a

significant increase in ALT and AST activities during hyper ammonia compared to that of clinically normal ones. These results were in consistence with those reported by **Mehra et al., (2005)** who attributed this increase to the degenerative changes in hepatic cells due to ammonia toxicity. The estimated levels of uric acid and urea in sheep only showed high significant concentrations which agreed with that of **Seiler et al., (1989)**, **Gamal et al., (1991)** and **Mehra et al., (2005)**. These findings were attributed to the fact that ammonia of whatever origin combined into organic linkage resulting in consequent formation of urea and uric acid leading to secondary hyperureaemia. The non significant increase of creatinine was in harmony with the finding of **Kubesy (1987)**. The reported decrease in serum calcium levels could be attributed to the concomitant increase of blood ammonia nitrogen levels, which leads to decrease calcium mobilization out of bone mean while the decrease of serum magnesium could be attributed to increase of ruminal ammonia nitrogen levels leading to chelation of magnesium, (**Radostitis et al., 1994**). Our serum total protein results did not change. while Serum albumin recorded high significant levels, meanwhile serum globulin concentration decreased significantly. Therefore A/G ratio increased significantly, our protein findings are agreeable to those previously mentioned by **Seleem et al., (1985)** and **Mehra et al., (2005)**.

Excess ammonia in rumen is absorbed across

the ruminal wall and taken to the liver to be converted to urea. Blood urea is freely diffusible to milk and therefore milk urea nitrogen reflects the urea concentration in blood (**Dias-Da-Silva and Sundstol, 1986**).

Ammonia is produced by the break down of protein during tissue metabolism and it is very toxic. Digestion of protein in the rumen releases ammonia that can be used by rumen bacteria (**Pell (1995)**).

Our results revealed that the elevation levels of milk urea nitrogen and decrease levels of proteins and fat which could be due to the excess of ammonia absorbed by the rumen which must be converted into urea by the liver for detoxification. Thus for dairy cows there are two entry points which may elevate blood and milk urea, the first is rumen degradation of protein and the second is degradation of protein by tissues. **Arunvipas, et al., (2003)** Concentration of milk urea has been used to monitor protein metabolism in dairy animal and blood urea concentration are a good estimate of the nutritional and reproductive status of animals (**Roseler et al., 1993**). Earlier research described much about relationships between urea concentration in serum and milk and nutritional management (**Godden et al., 2001 and Kamoun et al., 2012**). High level of milk urea usually reflect inadequate synchronisation between the carbohydrates available to fermentation in the rumen and rates of degradable and undegradable protein in the rumen (**Gustafsson and Carlsson 1993**). The basis for milk urea as nutritional indicator is the high correlation between the milk urea and blood urea nitrogen due to the free diffusion of urea in the organic tissue (**Melendez et al., 2000**).

A high milk urea nitrogen value suggests that energy is being utilized to convert ammonia to urea and is being diverted from milk production, animals with milk urea nitrogen values over 20 mg/dl will produce seven lb less milk (**Atzori et al., 2009**).

Animals have to expend 2 Mcal or more of energy to excrete the excess urea through the

urine. Thus excretion of excess urea is an energy requiring process. Excess concentration of urea in blood is believed to have detrimental effects on milk production, reproduction efficiency, embryo survivability and immune function **Ishler V (2008)**.

Urea also is an end product from tissue metabolism when the animal's tissues are energy deficient, protein are broken down within the animal's body to supply an energy source (**Faust and Kilmer 1998**).

Decreases in total protein, fat and total solid in milk may be attributed to malnutrition, leading consequently into emaciation (**Ratriguezl et al., 1997**).

In conclusion, the present investigation of deleterious effects of high ammonia percentage on productivity of sheep and goats, showed rapid emaciation and drop in milk production, changes in hematological and biochemical parameters. Also there were changes in milk constituents of affected animals.

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