A-16: N-Methyl nitroso urea (MNU) induced mammary gland tumours in the Sprague Dawley rats -a histopathological study

V Navaratne[1], B R R N Mendis[1], N V I Ratnatunga[2]
([1]Dept. of Oral Pathology, Faculty of Dental Science, Univ. of Peradeniya
[2]Dept. of Pathology, Faculty of Medicine, Univ. of Peradeniya)

Spontaneous and induced tumours of the rat mammary glands are widely used as experimental models in the study of human breast tumours. Many strains of rats develop spontaneous mammary tumours and also respond to different carcinogens by developing mammary tumours. The objective of this study is to monitor the effects of MNU which is a chemical carcinogen, on rat mammary glands. Studies have been reported in which MNU induced mammary tumorigenesis is seen in rats. This study describes the morphology and the histopathology of epithelial, fibroepithelial and connective tissue mammary glands tumours induced by MNU in the Sprague Dawley rats.

A single intravenous injection of chemical carcinogen N-Methyl nitrosourea (MNU) was administered to 110 (55 male and 55 female) 30 day old randomly bred Sprague Dawley rats. MNU is a N-nitroso compound which belongs to the group of non aromatic organic compounds. The chemical was dissolved in isotonic saline and a single dose of freshly prepared carcinogen 70 mg/kg body weight was injected to the tail vein of every rat. Matched control rats (n=62, 31 males and 31 females) were given the same volume of solvent only, by the same route. Animals were housed in plastic cages according to sex and fed a standard diet, prima broiler starter and water. The rats were weighed once a week. Rats were killed with an overdose of ether when they were obviously ill, moribund or otherwise 2-2½ years after the injection. At necropsy all macroscopically visible lesions were dissected, weighed, fixed in 10% formal saline and processed for histological examination by light microscopy. Sections from selected lesions were routinely stained with Haematoxylin and Eosin. Specific staining methods were adapted when necessary, to help with the histopathological diagnosis. Control animals were killed at the end of the experiment and processed in the identical manner.

Data from 49 treated (11 males 20% and 38 females 69%) rats who developed mammary tumours were available for evaluation. These tumours developed within a period of 2 months to 2 years after commencement of the MNU treatment. Tumours were either single or multiple ranging from 1-6 tumours/rat.The size varied between 1-13 cm in length and width. The tumours were localized in the regions of the mammary glands, arising from the right and left sides of upper cervico-thoracic mammary glands or from the lower abdomino-inguinal glands. Three main types of tumours: Epithelial 26%. Fibro-epithelial 66% and Connective tissue 8% were classified histologically. Majority of the tumours were benign fibro-adenomas which belong to the fibro-epithelial group. Macroscopically they were whitish tumours and were firm and rubbery in consistency. In the benign epithelial category adenomas, lactating adenomas and intraductal papillomas were found.

Malignant epithelial tumours such as ductal papillary carcinomas and ductal comedo carcinomas were also diagnosed. Macroscopically the malignant tumours were soft and fleshy growths. There were areas of necrosis, haemorrhage, ulceration and invasion into the neighbouring connective tissue and muscle tissue. The tumours of the malignant epithelial category were found only in the female rats.

Mammary glands tumours were induced in (35% females and 10% males) Sprague Dawley rats within a period of 2 months to 2 years after administering MNU. The sex difference was statistically significant ($p<0.01$). The mean latent period was 14 months. As in other studies tumours originated more often from the upper cervico thoracic mammary glands. Compared to other studies the latent period for the development of tumours was longer in males than in females. This difference was statistically significant ($p<0.05$)

Induced tumours comprise a range of histological types from benign epithelial to fibroepithelial and connective tissue tumours. There were also malignant epithelial tumours. Metastasis to distant organs was not obvious.

Table 1 Incidence of mammary tumours

	Number of rats					
	MNU			**Control**		
	Males	**Females**	**Total**	**Males**	**Females**	**Total**
Number injected	55	55	110	31	31	62
Number developed tumours	11 (20%)	38 (69.1%)	49 (45%)	3 (9.7%)	5 (16.1%)	8 (12.6%)
Number developed single tumours	5 (45%)	11 (29%)	16 (33%)	2	2	4
Number developed multiple tumours	6 (55%)	27 (71%)	33 (67%)	1	3	4

The difference between treated and control rats is statistically significant ($p<0.01$).

Table 2 Histopathology of mammary tumours

Histopathology	Number of Tumours			
	Male	Female	Total	%
Epithelial tumours				
Benign				
Adenomas	0	1	1	
Lactating	0	3	3	
Intraductal papillomas	1	0	1	
				26%
Malignant				
Ductal papillary carcinomas (invasive)	0	2	2	
Ductal comedo carcinomas (invasive)	0	3	3	
Fibroepithelial tumours				
Benign	2	23	25	66%
Fibroadenomas				
Connective tissue tumours				
Benign	1	2	3	8%
Fibromas				

Financial assistance by NARESA on Research grant RG/92/M/3 is acknowledged.