1- Effect of 6Gy gamma irradiation on chromosomal aberrations from Bone marrow cells of male albino rats:

In the present work, the side effect of irradiation was carried out.

- **Non exposure group (control):**

An attempt was made to get an idea about the chromosomal pattern of the rat. Therefore, as a starting point, metaphase chromosomes of the rats should be properly identified. For each rat, 50 metaphases were selected at random for microscopic examination. All of them had 42 chromosome per metaphase.

The chromosomes for each metaphase were karyotyped with regards to their length and position of centromere which taken as a basis for comparison with any deviation, which might be, occurred in the treated groups. The diploid number of chromosomes was found to be 42 per metaphase, (Fig, 2).

On Karyotyping, the autosomes did fit to the classification previously (Fig, 3).

According to **(Hungerford & Nowell, 1963)** The diploid number for all the strains was 42 chromosomes. They include 40 autosomes and 2 sex chromosomes. They classified the autosomes of the 3 strains in the following order:

Pair No. 1: The longest autosomes with sub terminal centromeres.

Pair No.2: The two longest autosomes with terminal centromeres.

Pair No.3: Satallited autosomes, with subterminal centromers. They are shorter than the autosomes of pair 1 and 2

Pair No. 4-10: Relatively large autosomes with terminal centromers, and gradually decreasing in length .

Pair No.11: Shorter than the mean length of group 4-10, but with subterminal centromere.

Pair No.12: Similar No.11 in length, with a subterminal centromere.

Pair No. 13: Shorter than 11 and 12 it has subtermiral centromere.

Pair No. 14-18: About the mean length of 11 and 12 but with median centromer.

Pair No. 19-20 : metacentric autosome, but shorter than autosomes of group 14-18.

The sex chromosome, however, furnished the main difference among the 3 strains; the X-chromosome presented in two forms, in lewis and wister strains. Once of these forms was an X with subterminal centromere and about the same size of autosome pairs number 3. The other form was a chromosome with a terminal centromers and about the same size as autosomes of group 4-10. The BN strain had one type of the X-chromosome. It was the terminal centromere. The Y-chromosome also present in two types. Once of them was a small acrocentric chromosome slightly larger than chromosome number 13. it was included in group 4-10 and could not be individually identified .

The chosen metaphases printed using suitable enlargements. One or two slides was chosen for each sample and 50 metaphases were examined randomly for chromosomal a berrations for each rat.

A search carried out for the chromosomal aberrations, which were either

(1) Structural chromosomal aberrations in the form of: chromosome breaks, gaps, dicentric, ring chromosomes and exchange figures.

(2) Numerical chromosomal aberrations in the form of: Aneuploidy and polyploidy.

The total chromosomal aberrations spotted on 23 metaphases from 300 metaphases. Occasionally, some sort of deviation in either the number or morphology of chromosome could be speculated. (Table, 1) The observed unusual findings included morphological findings reached to 5 occasions from 300 occasions mainly dicentric chromosomes, exchange figures and acentric fragment (Fig, 4). Numerical variations presented in the form of metaphase with a reduced number of chromosomes (Aneuploids) or metaphases with highly increased number of chromosomes (Polyploidy) (Fig, 4).The total numerical aberrations reached to 18 occasions from 300 occasions. However, such numerical or structural deviations were only randomly speculated.

- **Exposure group:**

The data obtained showed a marked rise in frequency of numerical and structural chromosomal aberrations after one, third, seventh, fifteen and thirty days post irradiation with 6Gy gamma rays.

After one day post irradiation with 6Gy of gamma ray (Table, 2) , the numerical aberrations were increased than structural aberrations, the numerical aberrations reached 166 occasions versus 18 from the control group.

These metaphases were presented with variable numbers of chromosomes. However, occasionally metaphases with a particular chromosomal number occurred in a suspicions frequency. Metaphases with hypodiploid number of chromosomes were shown up in 134 occasions (Table, 2 & Fig, 5), hyperdiploid numbers of chromosomes were shown up in 6 occasions. However, polyploid reached 26 occasions (Fig, 6).

Structural abnormalities were also much more numerous. Occasion of Dicentric chromosome, Ring, Exchange figures and acentric fragments were obvious (Fig, 7). Mitotic index which represents number of metaphases per number of non dividing cells that reach 2.3% after one day of radiation (Table, 7).

The numerical aberrations decreased from 166 occasions after the first day to 107 occasions (Table, 3). Moreover, the structural aberrations increased from 70 occasions after the first day to 78 occasions after the third day. The most structural aberrations were dicentric and acentric fragment while ring and exchange are similar in number, other aberrations such as deletion and breaks not spotted (Fig, 8). The percentage of total aberrations decreased from 78.66% after first day to 61.66% after the third day post-irradiation (Fig, 9a, b).

In addition, the same chromosome from different metaphases was hit dicentric aberration (Fig, 8).

Metaphases with hypodiploid number of chromosomes were shown in 80 occasions less than 134 after one day post-irradiation. Several metaphases had the same hypodiploid number of chromosomes (Fig, 10). Also mitotic index reach 2.7% after three day of radiation (Table, 7).

After seventh day post-irradiation, the numerical aberrations and structural aberrations were decreased. (Table, 4).

Data tabulated in (Table, 4), illustrated that the numerical aberrations reached up to 102 after the seventh day post-irradiation. Similarly the most numerical aberrations were hypodiploid (Fig, 11). However, the structural aberrations decreased than the other aberrations after the first and the third day post-irradiations, so that the structural aberrations reached to 57 occasions after the seventh days post-irradiation and the most structural aberrations were exchange figures and acentric fragment followed by dicentric (Fig, 12).

The percentage of total aberrations decreased from 61.66% after third day to 53.3% after seventh day post-irradiation. However, the high percentage was noted after the first day post irradiation as in (Table, 7 & Fig, 13a).

Also, the percentage of mitotic index was decreased and reached to 1.7%. After seventh day post-irradiation. After fifteen and thirty day post irradiation the numerical aberrations and structural aberrations were decreased than the other obtained data at previous intravel time of exposure as shown in (Table, 5 & 6). The numerical aberrations reached 72 after fifteen day and 51 after thirty day post-irradiation. However, the

structural aberrations decreased than the other data obtained, so that the structural aberrations reached 42 after fifteen day and 18 after thirty day post-irradiation while, the percentage of total chromosomal aberrations after the fifteen and thirty days reached to 38% and 23% respectively. Mitotic index after fifteen and thirty day post radiation reach 3% and 1.7% respectively histogramically (Fig, 13b & 14a).

The percentage of total chromosomal aberrations after the first day and the thirty day post irradiation reached 91.6% and 23% respectively, However, the high percentage was noted after the first day post-irradiation as in (Table, 7 & Fig, 14b).

While, the percentage of numerical chromosomal aberrations after the first, third, seventh, fifteen and thirty day post irradiation reached nearly 55.33%, 35.66%, 34% , 24% and 17% respectively (Table, 7 & Fig 14b).

The percentage of structural aberrations after the first, third, seventh, fifteen and thirty day post irradiation reached nearly 23.3% , 26%, 19%, 14% and 6% respectively (Table, 7 & Fig, 14b).

A total chromosomal aberrations were very highly significant after the first, third, seventh, fifteen and thirty days post irradiation as compared to control group (Table, 7).

Numerical aberrations were very highly significant after the first, third, seventh, fifteen and thirty days post-irradiation as compared to control group.

Structural aberrations were very highly significant after the first, third, seventh and fifteen days, while it become highly significant at thirty day post-irradiation.

Table (1): Frequencies of chromosomal aberrations from bone marrow cells of male rats in control group.

No. of rats	No. of scored cells	Chromosomal aberrations											Total chromosomal aberrations
		Numerical aberrations				Structural aberrations							
		Aneuploidy		Polyploidy	**Total**	Diceneric	Breaks	Deletion	Ring	Exchange figures	Acentric fragment	**Total**	
		Hypo-diploid	Hyper-diploid										
R1	**50**	**4**	**0**	**0**	**4**	**1**	**0**	**0**	**0**	**0**	**1**	**2**	**6**
R2	**50**	**3**	**0**	**0**	**3**	**0**	**0**	**0**	**0**	**2**	**0**	**2**	**5**
R3	**50**	**0**	**0**	**0**	**0**	**1**	**0**	**0**	**0**	**0**	**0**	**1**	**1**
R4	**50**	**4**	**0**	**0**	**4**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**4**
R5	**50**	**3**	**0**	**1**	**4**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**4**
R6	**50**	**2**	**0**	**1**	**3**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**3**
Sum	**300**	**16**	**0**	**2**	**18**	**2**	**0**	**0**	**0**	**2**	**1**	**5**	**23**
Mean	**50**	**2. 66**	**0**	**0. 33**	**3**	**0. 33**	**0**	**0**	**0**	**0. 33**	**0. 16**	**0. 83**	**3 . 83**
%		**5. 33**	**0**	**0. 66**	**6**	**0. 66**	**0**	**0**	**0**	**0. 66**	**0. 33**	**1. 66**	**7 . 6**
S.E		**0. 61**	**0.0**	**0. 21**	**0. 63**	**0. 21**	**0.0**	**0.0**	**0.0**	**0. 33**	**0. 16**	**0. 40**	**0. 70**

Table (2): Frequencies of chromosomal aberrations from bone marrow cells of male rats irradiated with 6Gy after one day.

No. of rats	No. of scored cells	Chromosomal aberrations											Total chromosomal aberrations
		Numerical aberrations				Structural aberrations							
		Aneuploidy		Polyploidy	**Total**	Diceneric	Breaks	Deletion	Ring	Exchange figures	Acentric fragment	**Total**	
		Hypo-diploid	Hyper-diploid										
R1	**50**	**22**	**2**	**5**	**29**	**3**	**1**	**0**	**3**	**3**	**4**	**14**	**43**
R2	**50**	**20**	**3**	**3**	**26**	**3**	**0**	**1**	**0**	**3**	**5**	**12**	**38**
R3	**50**	**23**	**1**	**7**	**31**	**2**	**0**	**0**	**5**	**2**	**4**	**13**	**44**
R4	**50**	**23**	**0**	**3**	**26**	**1**	**0**	**0**	**1**	**5**	**4**	**11**	**37**
R5	**50**	**23**	**0**	**5**	**28**	**1**	**0**	**0**	**1**	**4**	**3**	**9**	**37**
R6	**50**	**23**	**0**	**3**	**26**	**2**	**1**	**0**	**2**	**2**	**4**	**11**	**37**
Sum	**300**	**134**	**6**	**26**	**166**	**12**	**2**	**1**	**12**	**19**	**24**	**70**	**236**
Mean	**50**	**23. 33**	**1**	**4. 33**	**27. 66**	**2**	**0. 33**	**0. 16**	**2**	**3. 16**	**4**	**11. 66**	**39. 33**
%		**44. 66**	**2**	**8. 66**	**55. 33**	**4**	**0. 66**	**0. 33**	**4**	**6. 3**	**8**	**23. 3**	**78. 66**
S.E		**0. 49**	**0. 51**	**0. 66**	**0. 84**	**0. 36**	**0. 21**	**0. 16**	**0. 73**	**0. 47**	**0. 25**	**0. 71**	**1. 33**

Table (3): Frequencies of chromosomal aberrations from bone marrow cells of male rats irradiated with 6Gy after third day.

No. of rats	No. of scored cells	Chromosomal aberrations											Total chromosomal aberrations
		Numerical aberrations				Structural aberrations							
		Aneuploidy		Polyploidy	**Total**	Diceneric	Breaks	Deletion	Ring	Exchange figures	Acentric fragment	**Total**	
		Hypo-diploid	Hyper-diploid										
R1	**50**	**9**	**2**	**5**	**16**	**5**	**0**	**0**	**4**	**2**	**3**	**14**	**30**
R2	**50**	**17**	**1**	**2**	**20**	**5**	**0**	**0**	**1**	**2**	**3**	**11**	**31**
R3	**50**	**11**	**0**	**2**	**13**	**3**	**0**	**0**	**4**	**4**	**2**	**13**	**26**
R4	**50**	**13**	**2**	**4**	**19**	**5**	**0**	**1**	**2**	**3**	**3**	**14**	**33**
R5	**50**	**15**	**1**	**2**	**18**	**4**	**0**	**0**	**2**	**3**	**4**	**13**	**31**
R6	**50**	**15**	**1**	**5**	**21**	**4**	**0**	**0**	**2**	**1**	**6**	**13**	**34**
Sum	**300**	**80**	**7**	**20**	**107**	**26**	**0**	**1**	**15**	**15**	**21**	**78**	**185**
Mean	**50**	**13. 33**	**1. 16**	**3. 33**	**17. 83**	**4. 33**	**0**	**0. 16**	**2. 5**	**2. 5**	**3. 5**	**13**	**30. 83**
%		**26. 66**	**2. 33**	**6. 66**	**35. 66**	**8. 66**	**0**	**0. 33**	**5**	**5**	**7**	**26**	**61. 66**
S.E		**1. 20**	**0. 30**	**0. 61**	**1. 19**	**0. 33**	**0.0**	**0. 16**	**0. 5**	**0. 42**	**0. 56**	**0. 44**	**1. 13**

Table (4): Frequencies of chromosomal aberrations from bone marrow cells of male rats irradiated with 6Gy after seven days.

No. of rats	No. of scored cells	Chromosomal aberrations											Total chromosomal aberrations
		Numerical aberrations				Structural aberrations							
		Aneuploidy		Polyploidy	**Total**	Diceneric	Breaks	Deletion	Ring	Exchange figures	Acentric fragment	**Total**	
		Hypo-diploid	Hyper-diploid										
R1	**50**	**14**	**0**	**3**	**17**	**3**	**0**	**0**	**1**	**4**	**3**	**11**	**28**
R2	**50**	**15**	**1**	**0**	**16**	**3**	**0**	**3**	**2**	**3**	**5**	**16**	**32**
R3	**50**	**14**	**0**	**2**	**16**	**3**	**0**	**0**	**2**	**1**	**2**	**8**	**24**
R4	**50**	**15**	**1**	**2**	**18**	**0**	**2**	**2**	**0**	**2**	**2**	**8**	**26**
R5	**50**	**14**	**1**	**4**	**19**	**2**	**0**	**0**	**1**	**2**	**3**	**8**	**27**
R6	**50**	**13**	**0**	**3**	**16**	**0**	**0**	**0**	**1**	**4**	**1**	**6**	**22**
Sum	**300**	**85**	**3**	**14**	**102**	**11**	**2**	**5**	**7**	**16**	**16**	**57**	**159**
Mean	**50**	**14. 16**	**0. 5**	**2. 33**	**17**	**1. 83**	**0. 33**	**0. 83**	**1. 16**	**2. 66**	**2. 66**	**9. 5**	**26. 5**
%		**28. 33**	**1**	**4. 66**	**34**	**3. 66**	**0. 66**	**1. 66**	**2. 33**	**5. 33**	**5. 33**	**19**	**53**
S.E		**0. 30**	**0. 22**	**0. 55**	**0. 51**	**0. 60**	**0. 33**	**0. 54**	**0. 30**	**0. 49**	**0. 55**	**1. 45**	**1. 40**

Table (5): Frequencies of chromosomal aberrations from bone marrow cells of male rats irradiated with 6Gy after fifteen days.

No. of rats	No. of scored cells	Chromosomal aberrations											Total chromosomal aberrations
		Numerical aberrations				Structural aberrations							
		Aneuploidy		Polyploidy	**Total**	Diceneric	Breaks	Deletion	Ring	Exchange figures	Acentric fragment	**Total**	
		Hypo-diploid	Hyper-diploid										
R1	**50**	**9**	**3**	**2**	**14**	**1**	**1**	**1**	**1**	**2**	**0**	**6**	**20**
R2	**50**	**8**	**0**	**0**	**8**	**3**	**0**	**1**	**1**	**3**	**1**	**9**	**17**
R3	**50**	**11**	**1**	**1**	**13**	**0**	**0**	**1**	**1**	**2**	**3**	**7**	**20**
R4	**50**	**10**	**0**	**3**	**13**	**3**	**1**	**0**	**1**	**3**	**1**	**9**	**22**
R5	**50**	**8**	**2**	**2**	**12**	**1**	**0**	**0**	**2**	**1**	**1**	**5**	**17**
R6	**50**	**10**	**0**	**2**	**12**	**1**	**0**	**0**	**1**	**2**	**2**	**6**	**18**
Sum	**300**	**56**	**6**	**10**	**72**	**9**	**2**	**3**	**7**	**13**	**8**	**42**	**114**
Mean	**50**	**9. 33**	**1**	**1. 66**	**12**	**1. 5**	**0. 33**	**0. 5**	**1. 16**	**2. 16**	**1. 33**	**7**	**19**
%		**18. 66**	**2**	**3. 33**	**24**	**3**	**0. 66**	**1**	**2. 33**	**4. 33**	**2. 66**	**14**	**38**
S.E		**0. 49**	**0. 51**	**0. 42**	**0. 85**	**0. 5**	**0. 21**	**0. 22**	**0. 16**	**0. 30**	**0. 42**	**0. 68**	**0. 81**

Table (6): Frequencies of chromosomal aberrations from bone marrow cells of male rats irradiated with 6Gy after 30 day.

No. of rats	No. of scored cells	Chromosomal aberrations											Total chromosomal aberrations
		Numerical aberrations				Structural aberrations							
		Aneuploidy		Polyploidy	**Total**	Diceneric	Breaks	Deletion	Ring	Exchange figures	Acentric fragment	**Total**	
		Hypo-diploid	Hyper-diploid										
R1	**50**	**5**	**0**	**3**	**8**	**0**	**0**	**0**	**2**	**1**	**2**	**5**	**13**
R2	**50**	**9**	**0**	**3**	**12**	**0**	**0**	**0**	**2**	**1**	**0**	**3**	**15**
R3	**50**	**7**	**0**	**0**	**7**	**0**	**0**	**0**	**1**	**1**	**0**	**2**	**9**
R4	**50**	**8**	**0**	**1**	**9**	**1**	**0**	**0**	**0**	**1**	**0**	**2**	**11**
R5	**50**	**6**	**0**	**0**	**6**	**2**	**0**	**0**	**1**	**2**	**0**	**5**	**11**
R6	**50**	**7**	**0**	**0**	**2**	**9**	**0**	**0**	**0**	**0**	**1**	**0**	**1**
Sum	**300**	**42**	**0**	**9**	**51**	**3**	**0**	**0**	**6**	**7**	**2**	**18**	**69**
Mean	**50**	**7**	**0**	**1. 5**	**8. 5**	**0. 5**	**0**	**0**	**1**	**1. 16**	**0. 33**	**3**	**11. 5**
%		**14**	**0**	**3**	**17**	**1**	**0**	**0**	**2**	**2. 33**	**0. 66**	**6**	**23**
S.E		**0. 57**	**0.0**	**0. 56**	**0. 84**	**0. 34**	**0.0**	**0.0**	**0. 36**	**0. 16**	**0. 33**	**0. 68**	**0.88**

Table (7): Comparison between the frequencies of chromosomal aberrations from bone marrow cells in male rats irradiated with 6Gy gamma-radiation.

Treatment	No. of Rats	No. of Scored cells 50/rat	Chromosomal aberrations									% of Mitotic Index
			Numerical aberrations			Structural aberration			Total aberrations			No. of metaphase / No of non-dividing cells
			No.	*%*	*Mean ± SE*	*No.*	*%*	*Mean ± SE*	*No.*	*%*	*Mean ± SE*	
Control	**6**	**300**	**18**	**6**	**3 ± 0.63**	**5**	**1.66**	**0.83 ± 0.16**	**23**	**7.66**	**3.83 ± 0.70**	**2%**
1st day	**6**	**300**	**166**	**55.33**	**27.66 ± 0.84*****	**70**	**23.3**	**11.66 ± 0.71*****	**236**	**78.66**	**39.3 ± 1.33*****	**2.3%**
3rd day	**6**	**300**	**107**	**35.66**	**17.83 ± 1.19*****	**78**	**26**	**13 ± 0.44*****	**185**	**61.66**	**30.83 ± 1.13*****	**2.7%**
7th day	**6**	**300**	**102**	**34**	**17 ± 0.51*****	**57**	**19**	**9.5 ± 1.45*****	**159**	**53**	**26.5 ± 1.40*****	**1.7%**
15th day	**6**	**300**	**72**	**24**	**12 ± 0.85*****	**42**	**14**	**7 ± 0.68*****	**114**	**38**	**19 ± 0.81*****	**3%**
30th day	**6**	**300**	**51**	**17**	**8.5 ± 0.84*****	**18**	**6**	**3 ± 0.68***	**69**	**23**	**11.5 ± 0.88*****	**1.7%**

*** Significant at P < 0.05** **** Significant at P < 0.01** ***** Significant at P < 0.001**

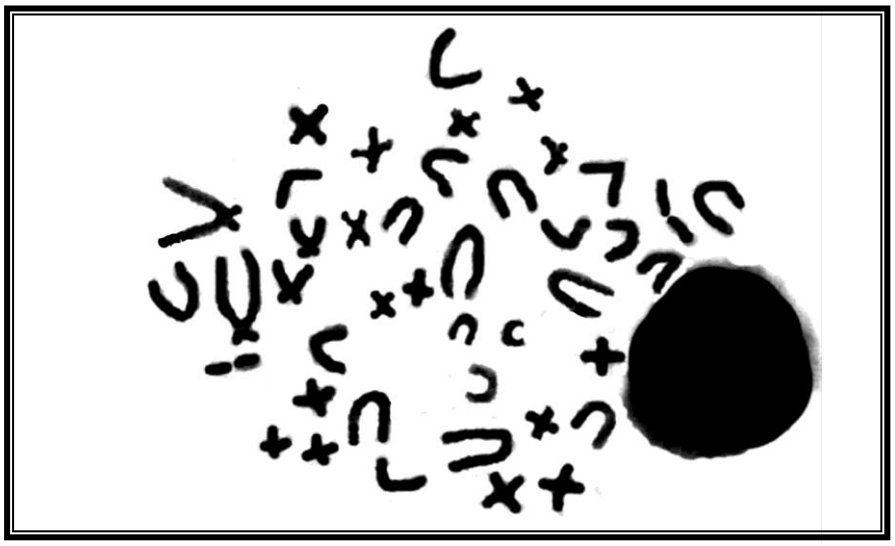


Fig. (2): Metaphase spread of untreated male albino rat (40 x y). Giemsa stain & X .1000.

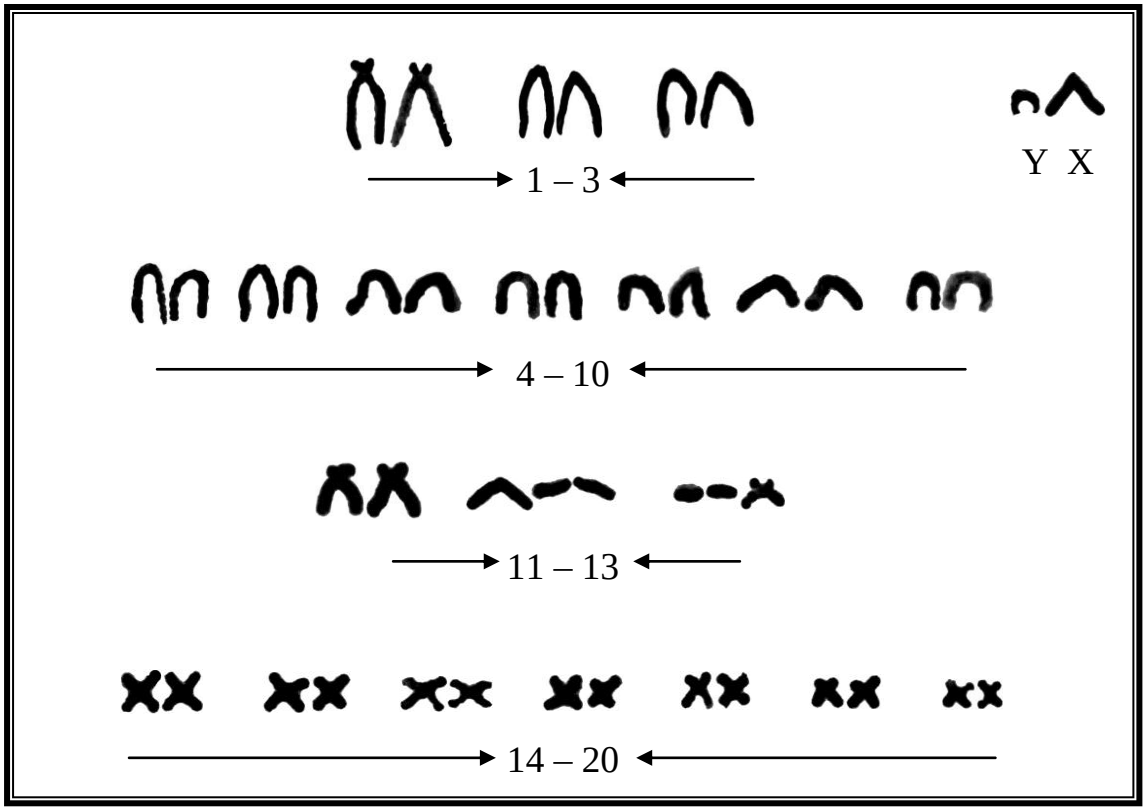


Fig (3): Karyotype of the up metaphase spread of untreated male albino rat & X .1000.

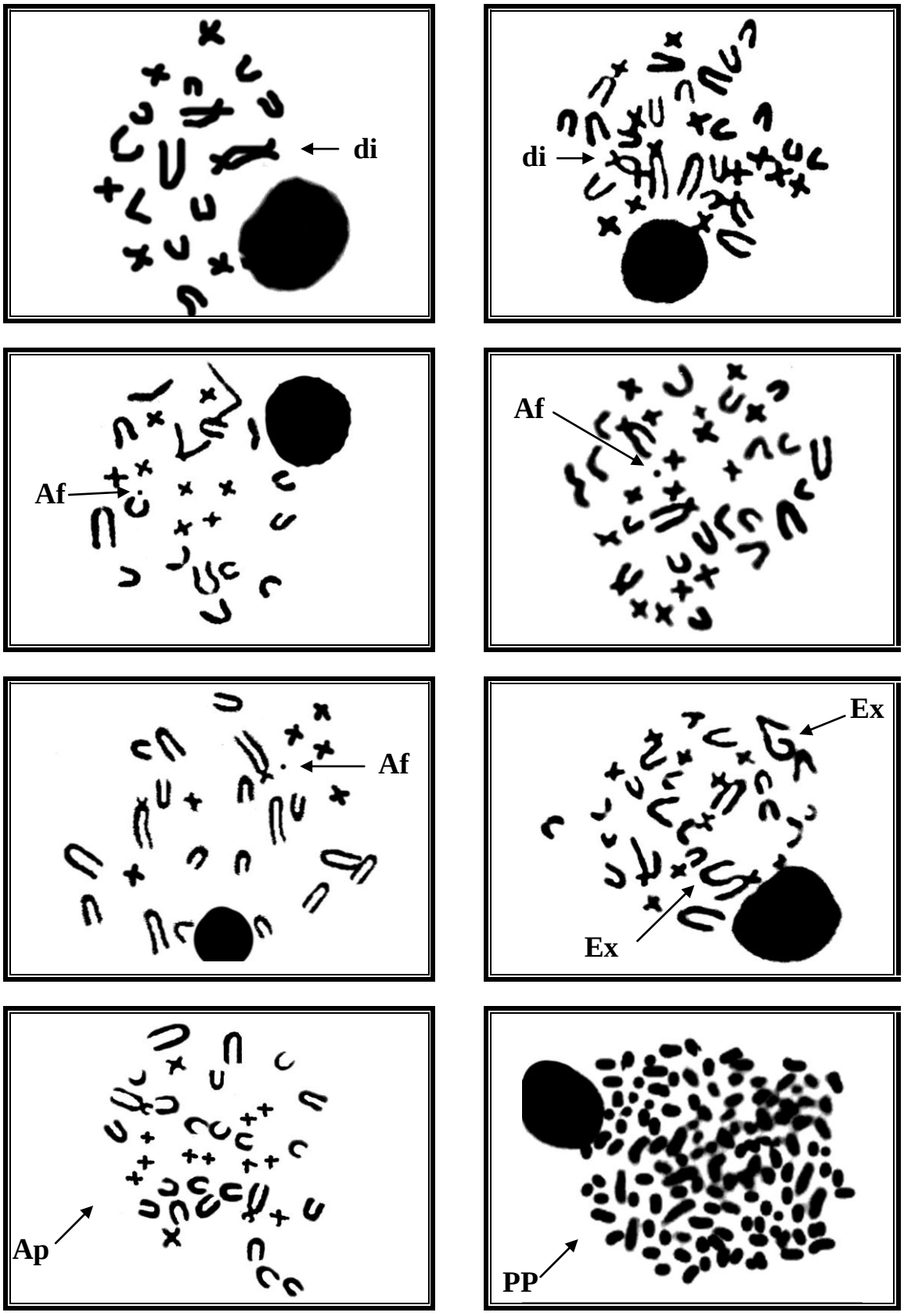


Fig (4): Metaphases with unusual structural and numerical chromosomes from control rat, dicentric (di), Exchange figure (Ex), Acentric fragment (Af), Aneuploidy (Ap), Polyploidy (pp).

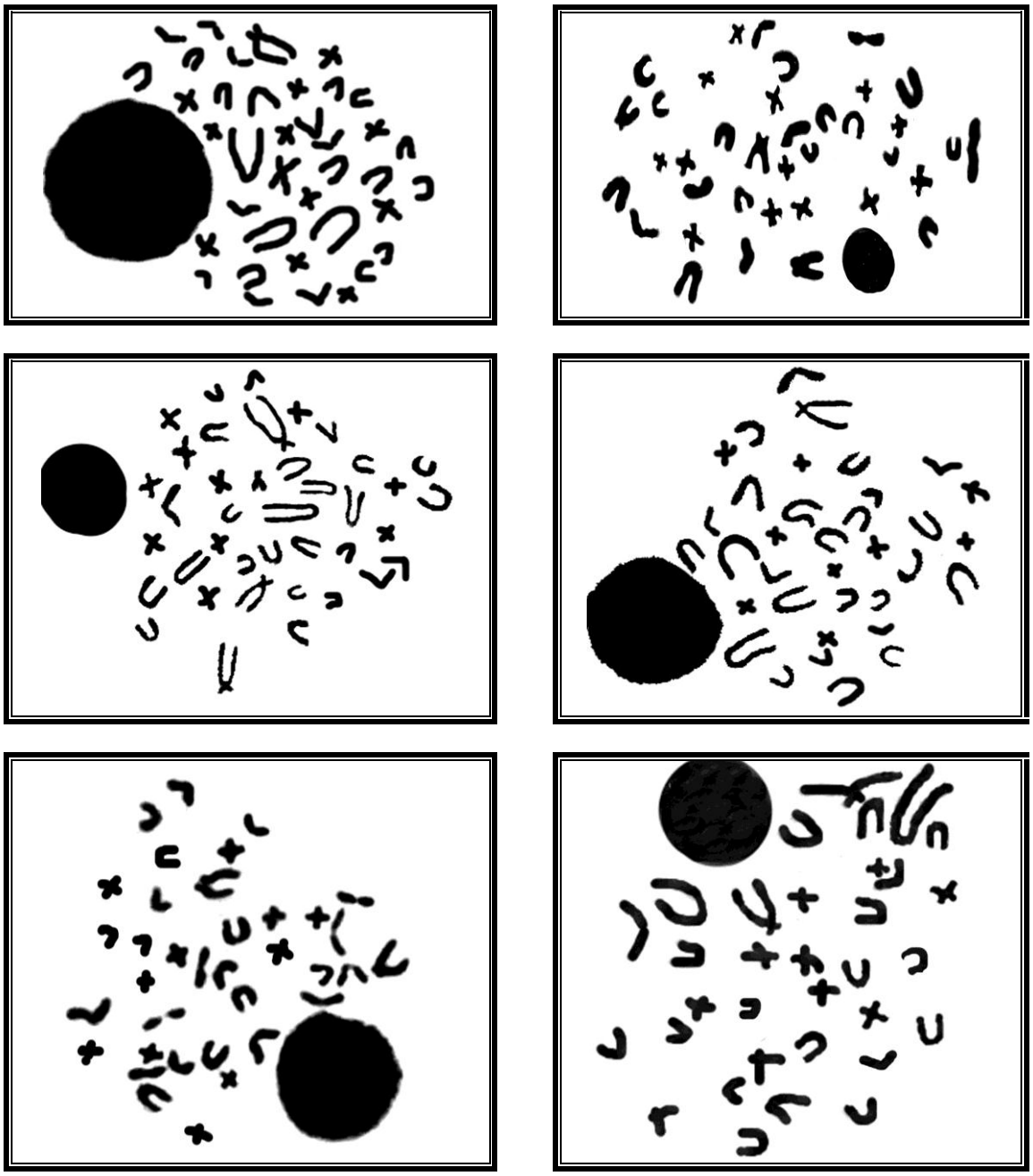


Fig (5): Metaphases with reduced number of chromosomes from bone marrow cells of rat exposed to irradiation after one day.

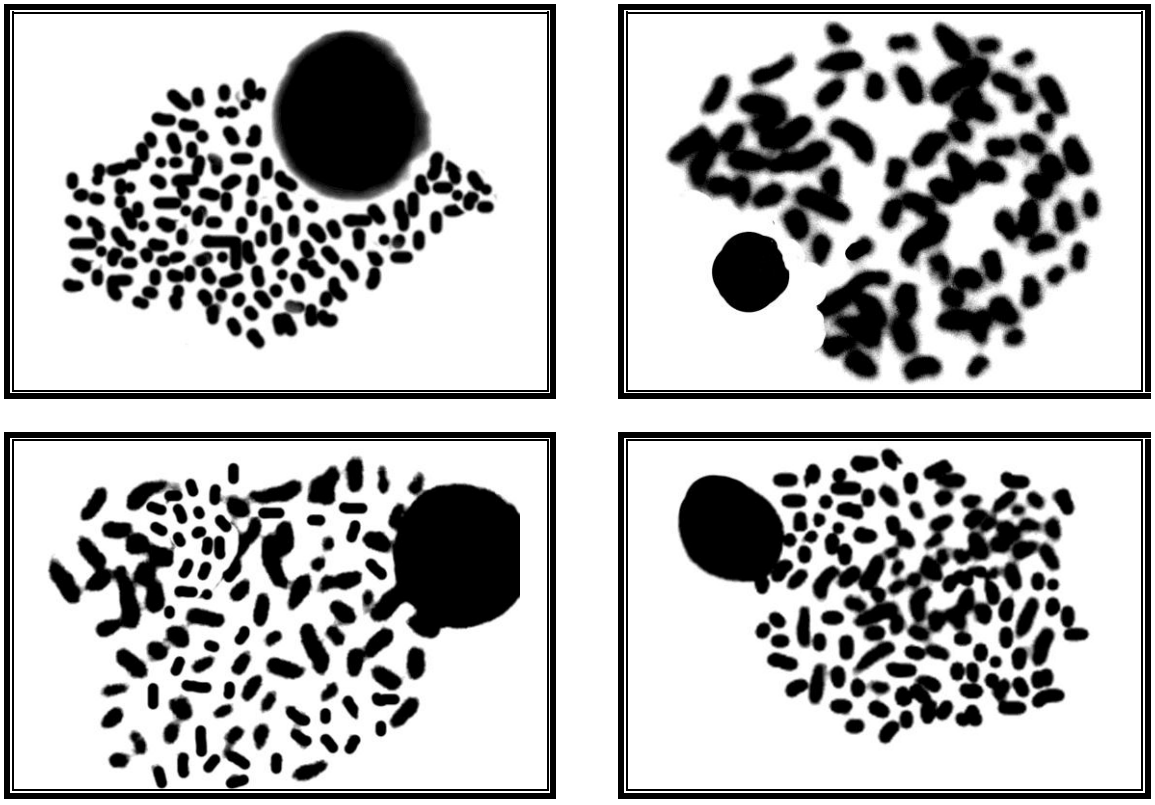


Fig (6): Metaphases with polyploidy chromosomes from bone marrow cells of rat exposed to irradiation after one day.

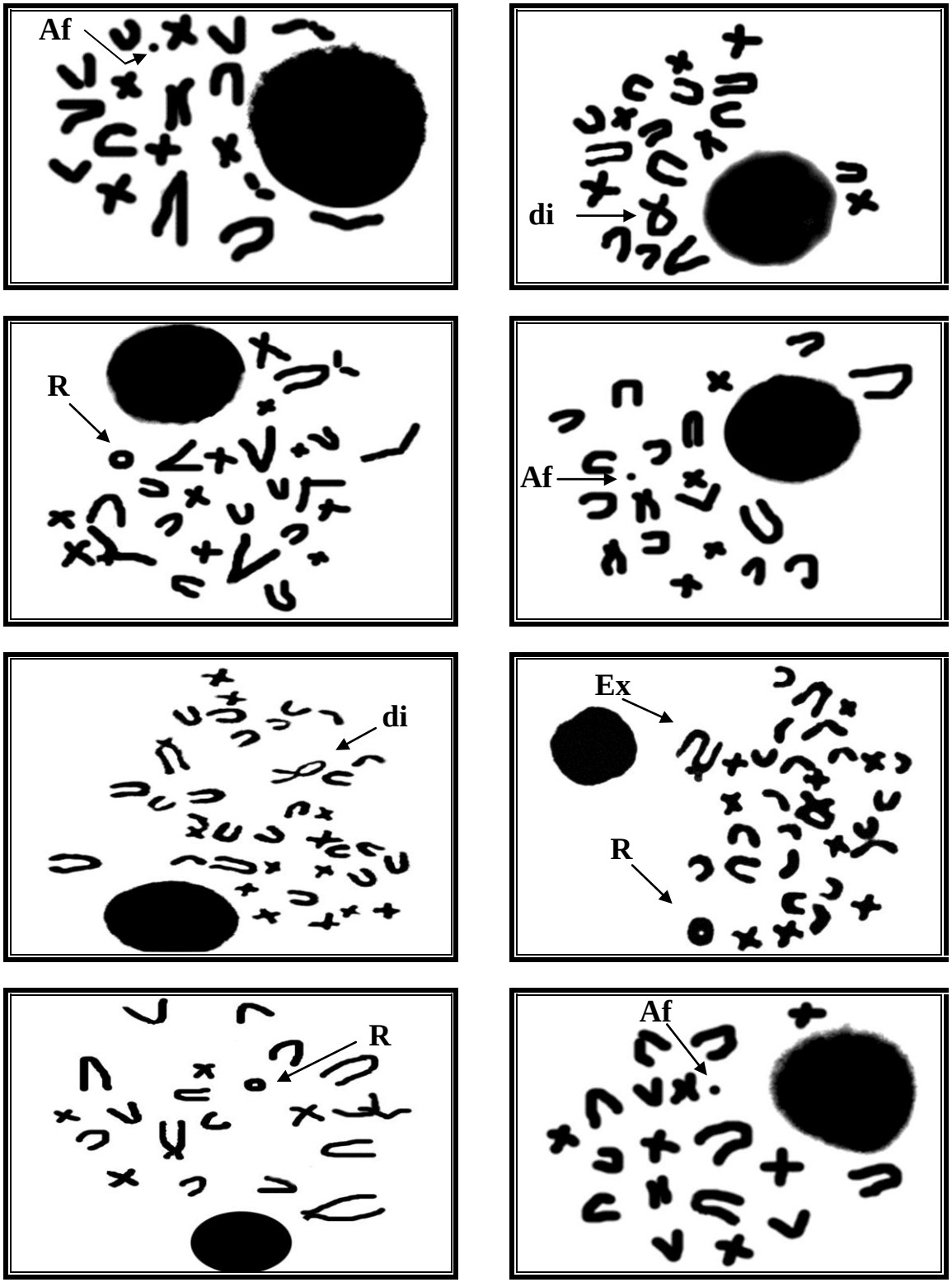


Fig (7): Metaphases with structural aberrations after one day of exposure to irradiation dicentric (di), Ring (R), Exchange figures (Ex), Acentric fragment (Af).

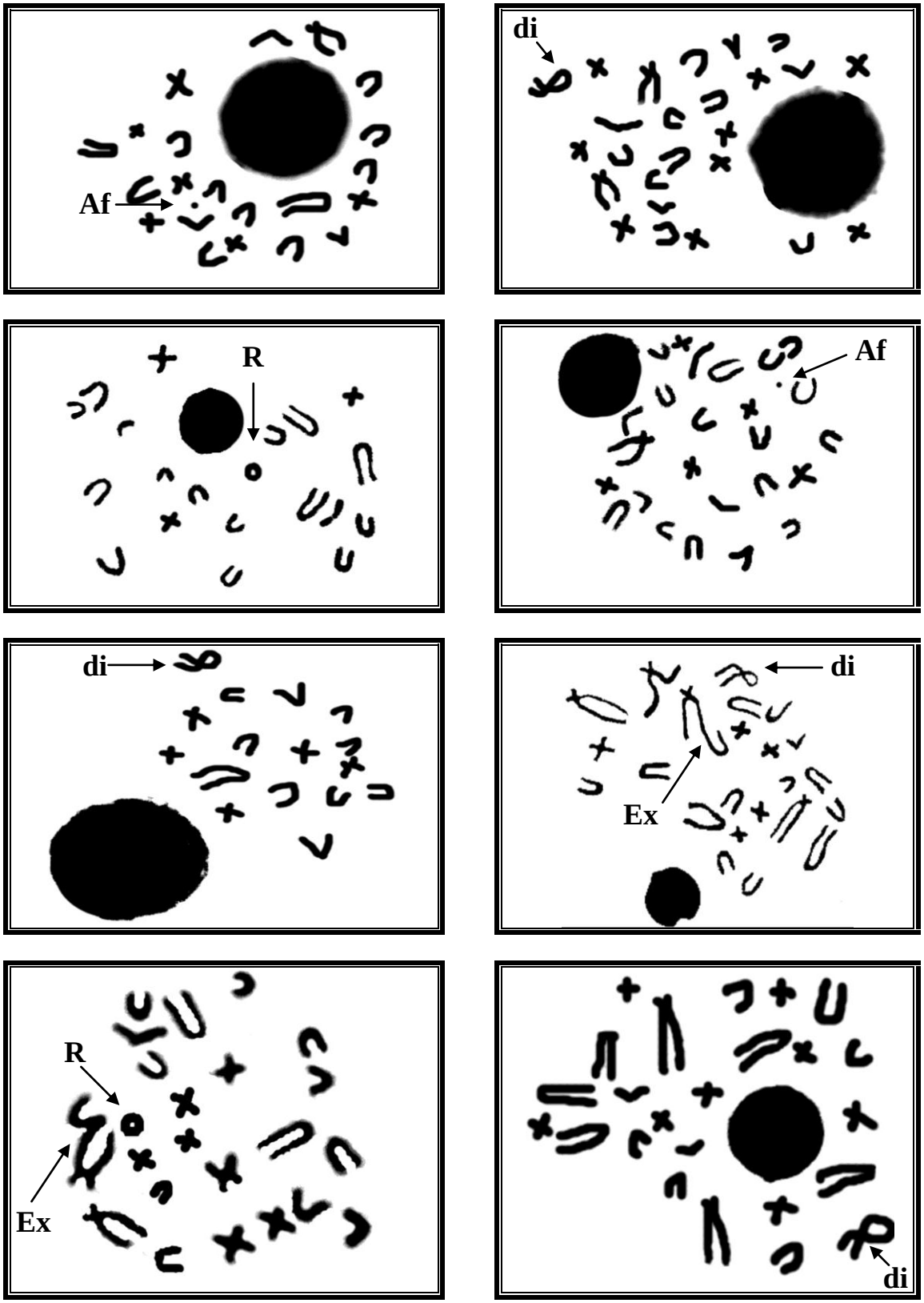


Fig (8): Metaphases with unusual structural chromosomes from rat after 3 day from exposed to irradiation dicentric (di), Exchange figure (Ex), Acentric fragment (Af), Ring (R).

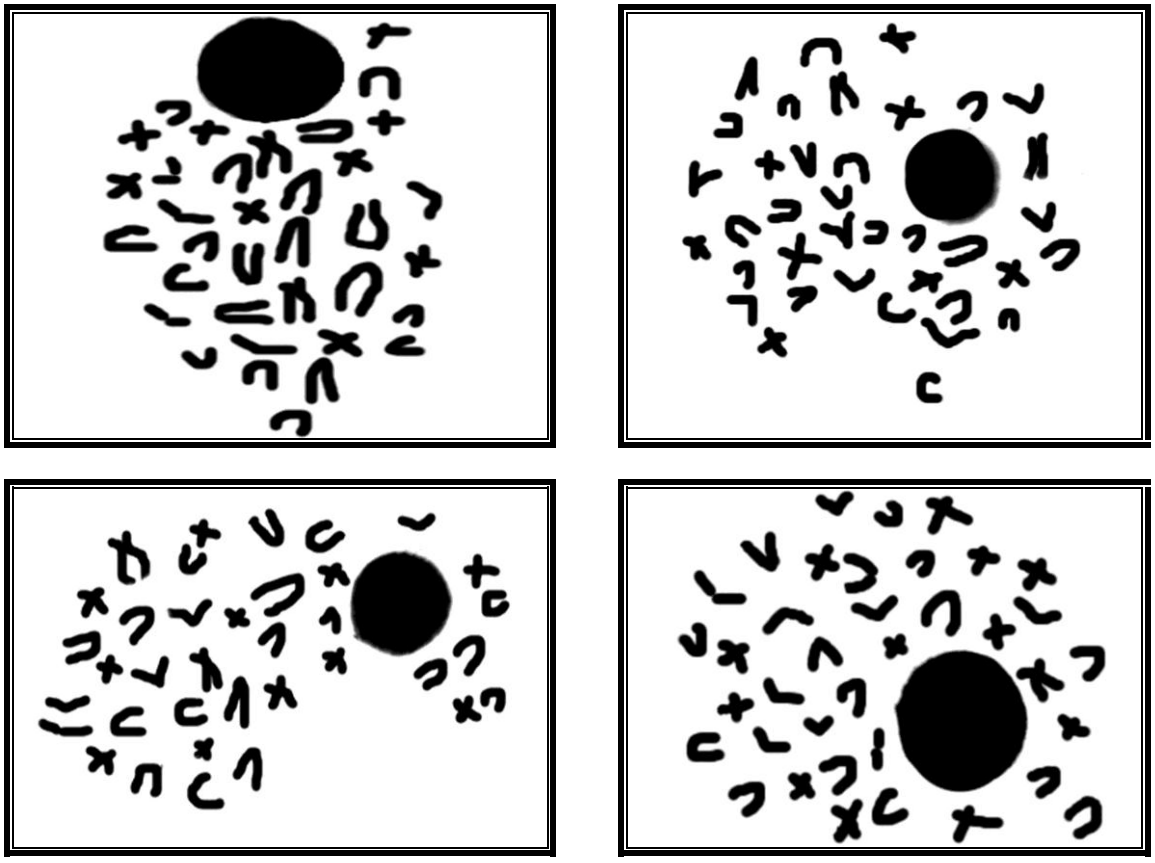


Fig (10): Several metaphases had the same hypodiploid number of chromosomes (36 chromosomes) after 3 day post-irradiation.

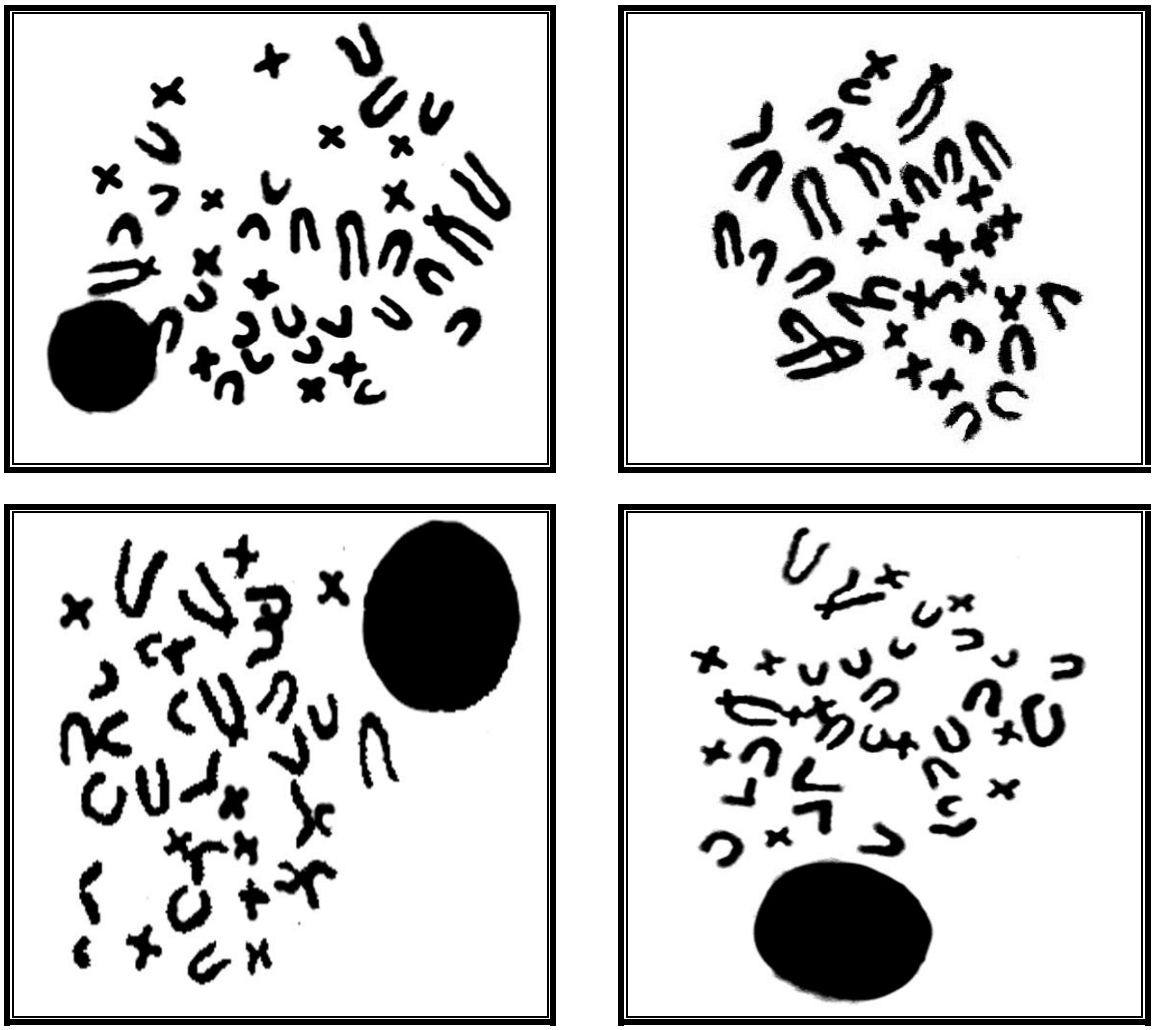


Fig (11): Metaphases had hypodiploid chromosomal number (variable number) from rat after 7 day post-irradiation.

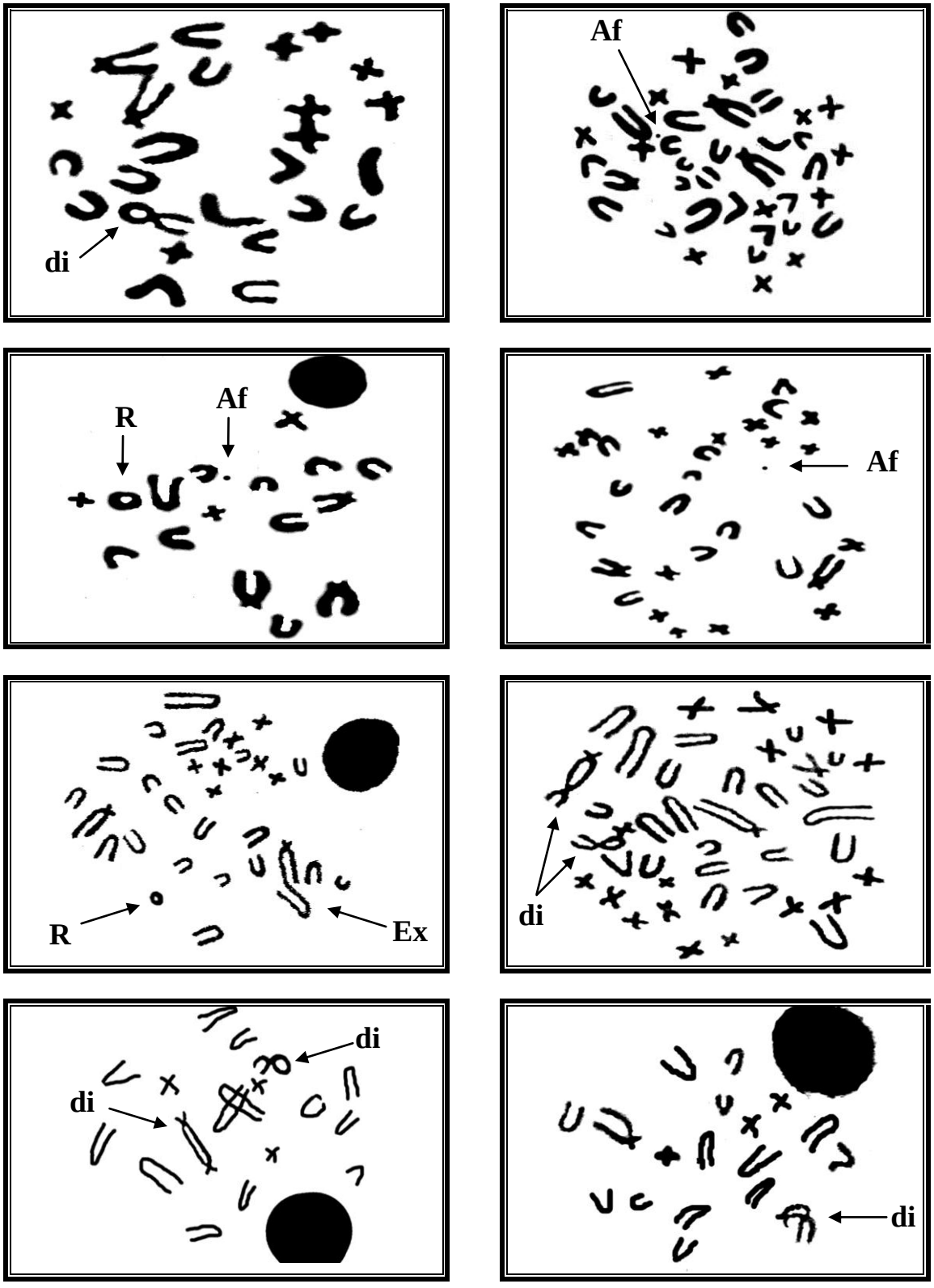


Fig (12): Metaphase had structural aberrations from rat after 7 days post-irradiation dicentric (di), Exchange figure (Ex), Acentric fragment (Af), Ring (R).

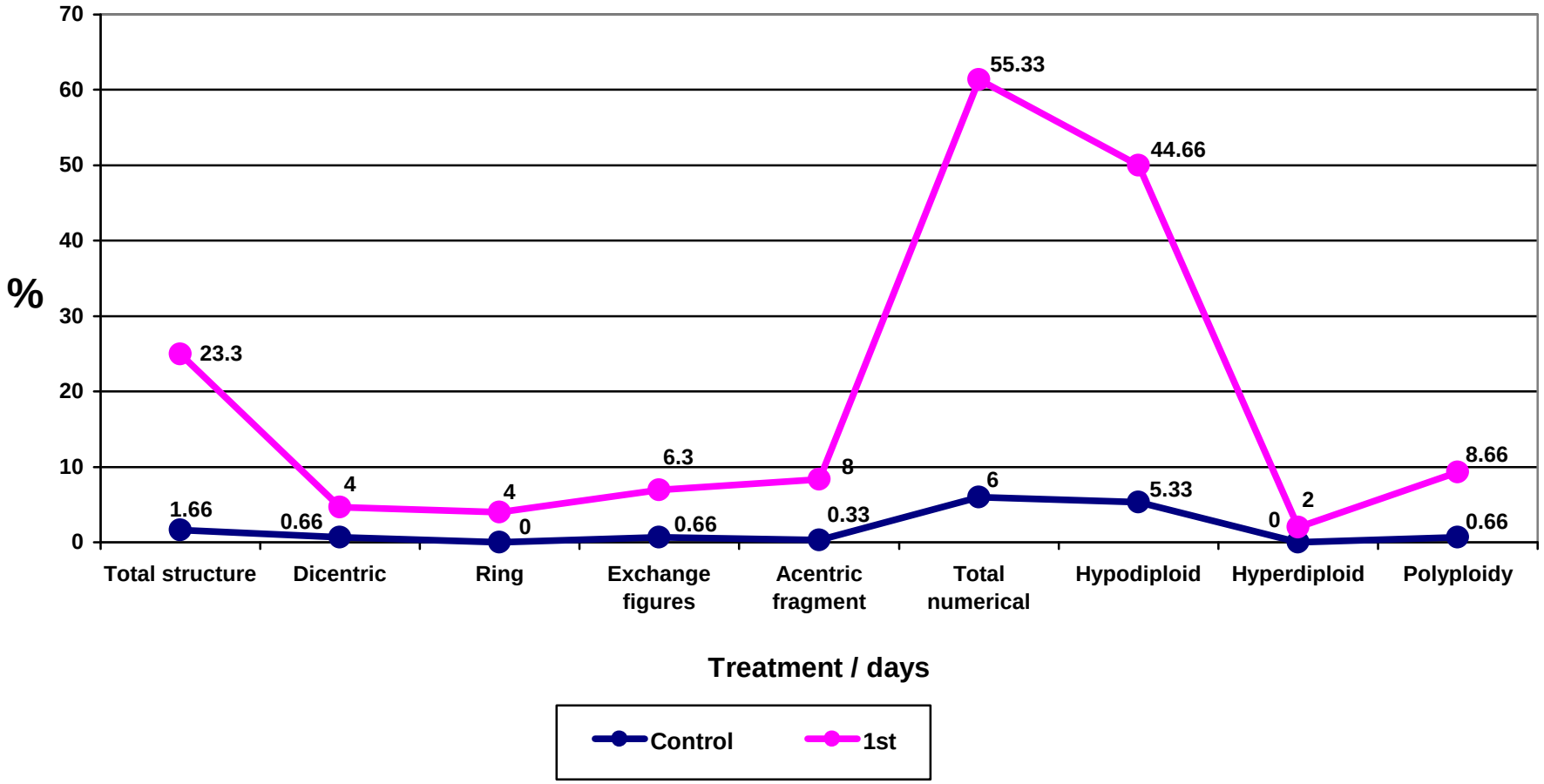


Fig (9a): The percentage of numerical type aberration and structural type aberration from bone marrow cells of male rat irradiated with 6Gy gamma radiation after one day.

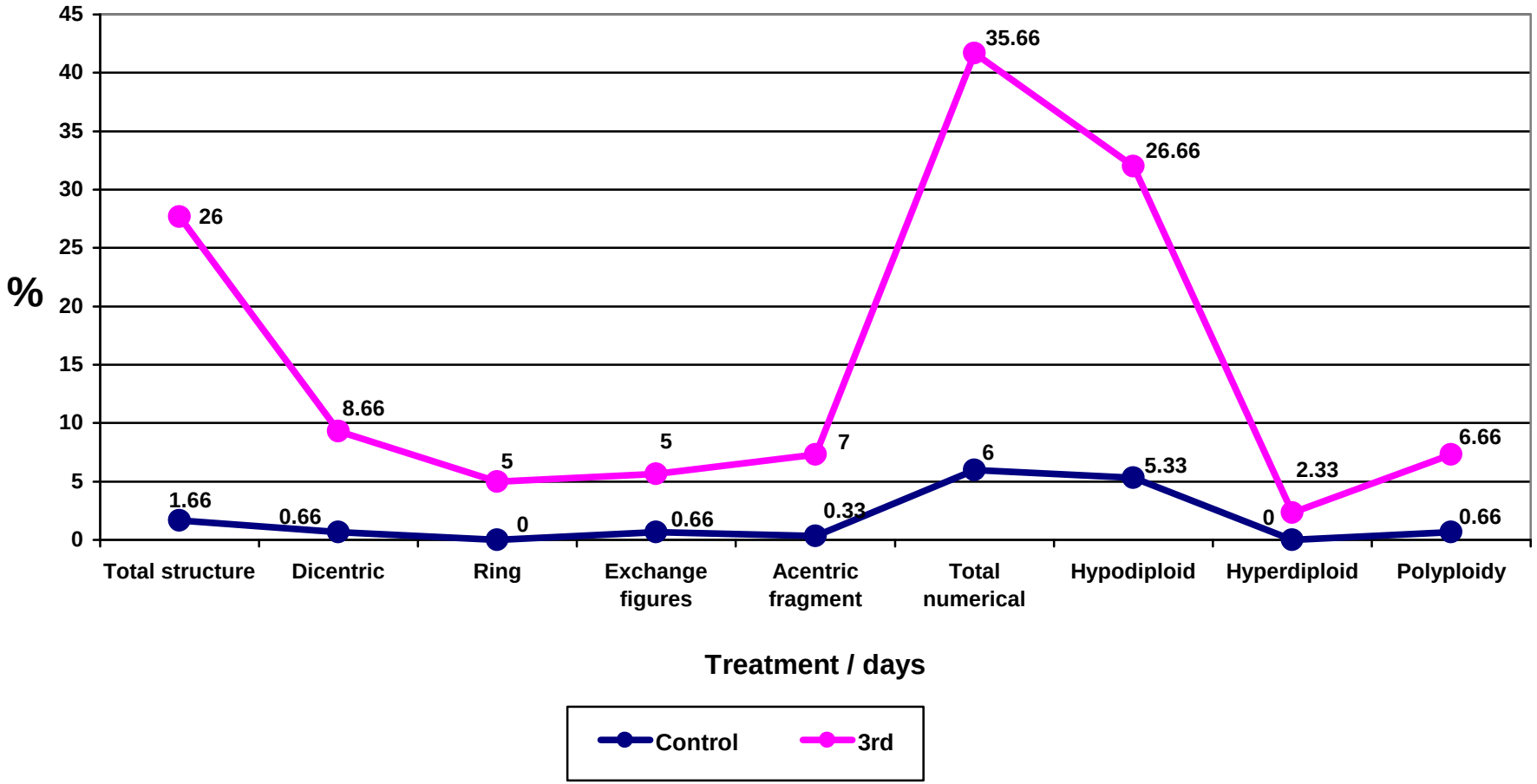


Fig (9b): The percentage of numerical type aberration and structural type aberration from bone marrow cells of male rat irradiated with 6Gy gamma radiation after third day.

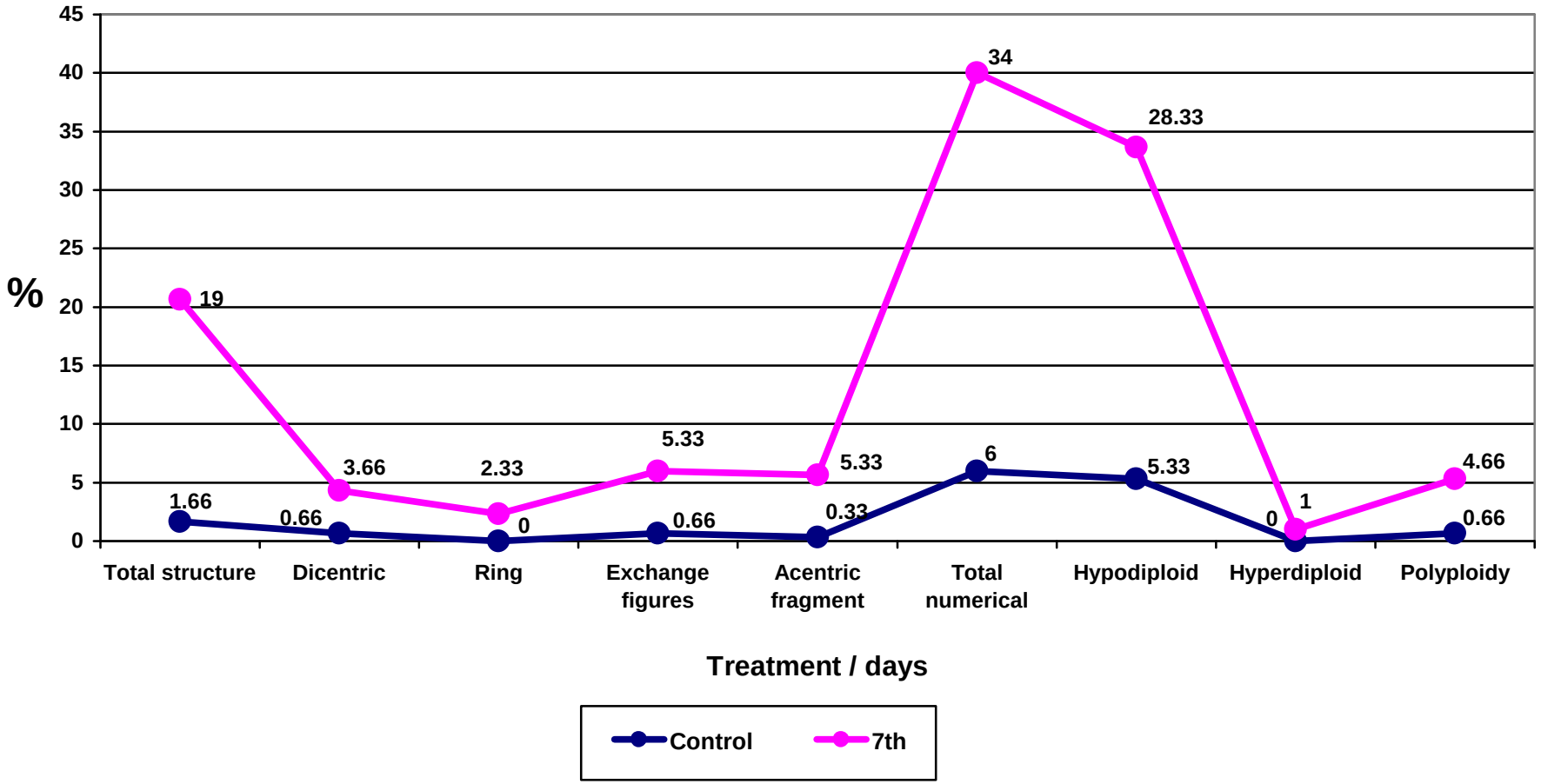


Fig (13a): The percentage of numerical type aberration and structural type aberration from bone marrow cells of male rat irradiated with 6Gy gamma radiation after seventh day.

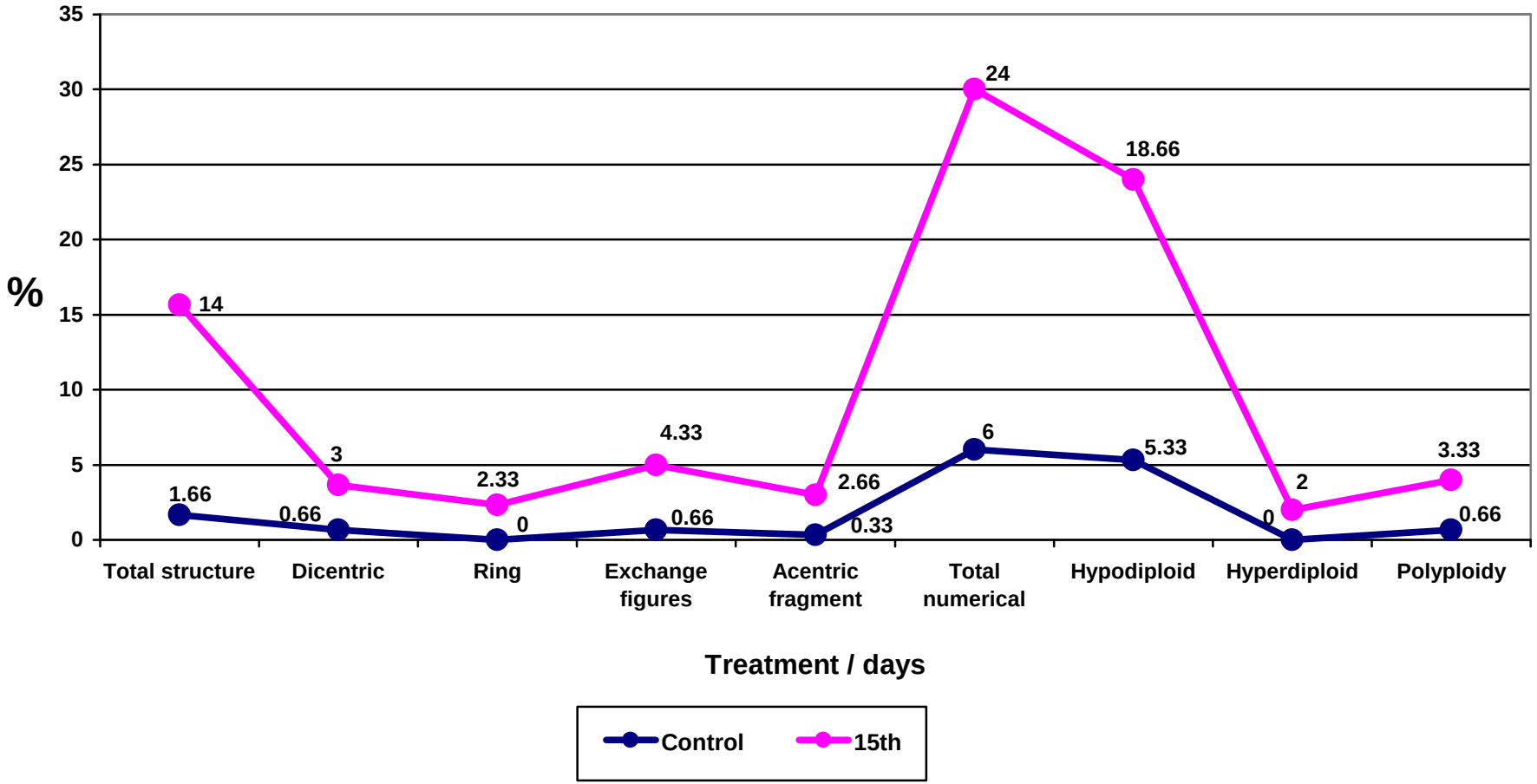


Fig (13b): The percentage of numerical type aberration and structural type aberration from bone marrow cells of male rat irradiated with 6Gy gamma radiation after fifth day.

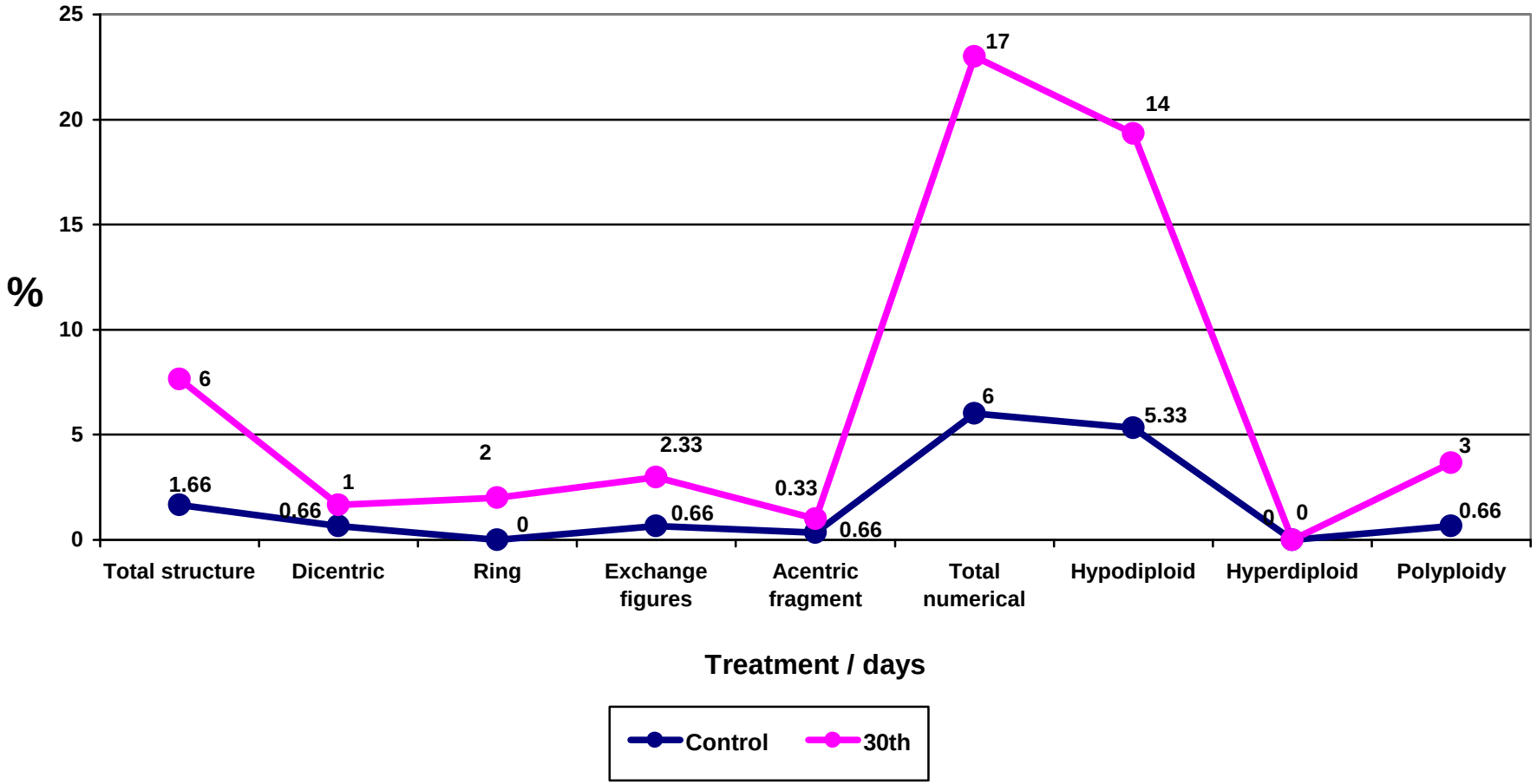


Fig (14a): The percentage of numerical type aberration and structural type aberration from bone marrow cells of male rat irradiated with 6Gy gamma radiation after thirty day.

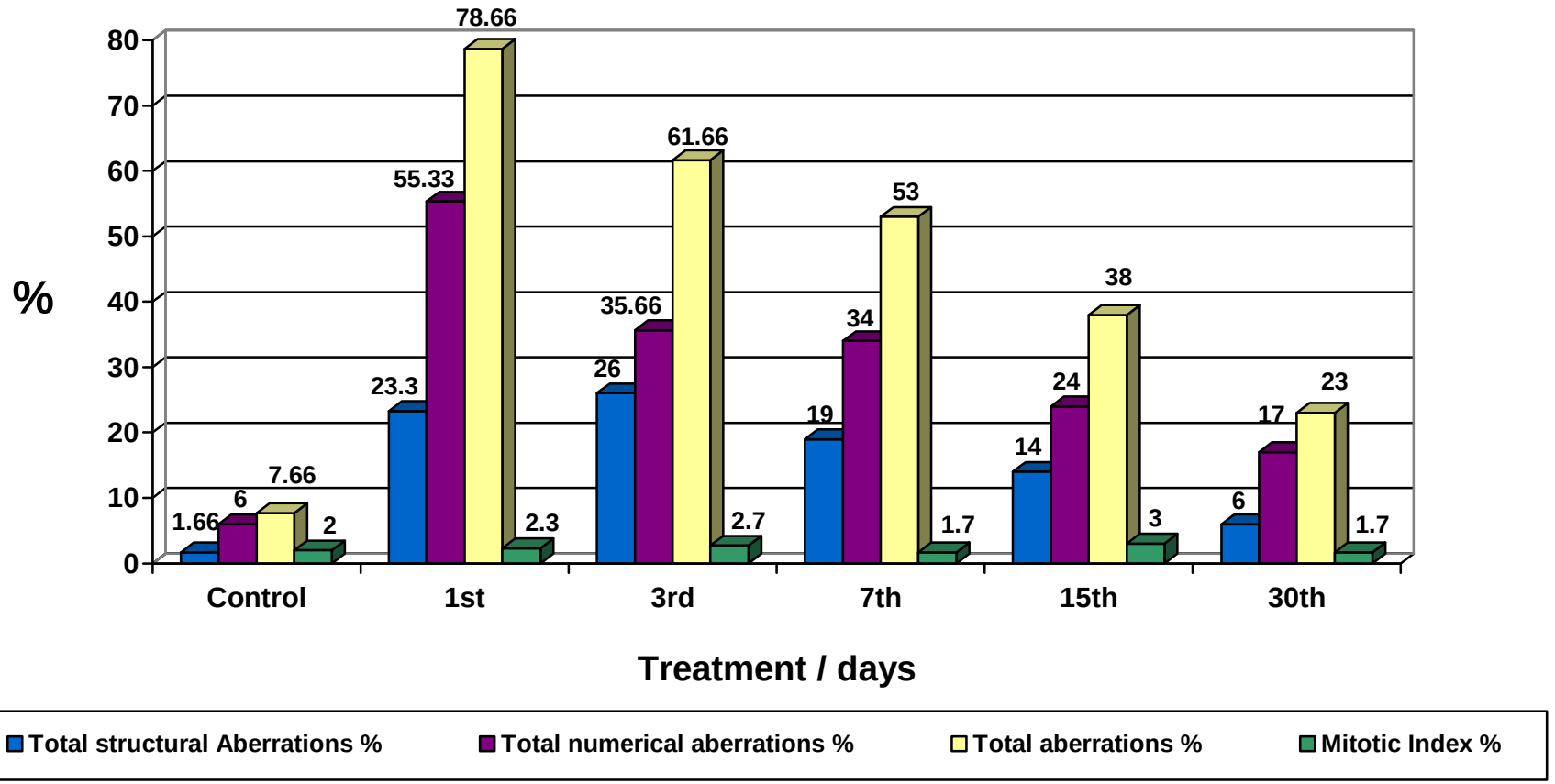


Fig (14b): Comparison between the percentage of structural, numerical, total aberrations and mitotic index from bone marrow cells of male rat irradiated with 6Gy gamma radiation.

Effect of 6Gy gamma irradiation on Micronuclei from Bone marrow cells of Rats:-

In the present work , the side effect of 6Gy gamma irradiation was carried out. The micronuclei may originate from a centric fragments (chromosome fragments lacking a centromere) or whole chromosomes which are unable to migrate with the rest of the chromosomes during the anaphase of cell division.

At least 1000 micronuclei polychromatic erythrocyte cells examined blindly are scored to assess the frequency of cells with one, two, or more than two micronuclei.

The frequency of MNPCEs in non treated condition was determined in 6000 bone marrow cells from the control group of 6 male rats (41 $\pm$ 1.5) out of each 1000 MNPCEs, had micronuclei (Table 8).

The cells are classified as single micronuclei, bi-micronuclei or multi-micronuclei (Fig, 15).

After one day post-irradiation, the number of micronuclei reached 1787 occasions as compared with 246 occasions at the control groups. Table (9). The single micronuclei, bi-micronuclei and multi-micronuclei on the first day reached 1586, 162, 39 respectively.

Also, the microneucleus was varied in size and shape, some micronucleus was small, other was large. While some micronuclei had round, linear and/or almond in shape (Fig, 16).

The percentage of total micronuclei aberrations after the first day post-irradiation reached nearly 29.78% while in the control groups it

reached 4.1%. In addition the bone marrow activity which represents the activity of bone marrow for formation of micronuclei and equal to Mean / 1000 reach 0.29783 after one day post-irradiation (Fig, 17).

After third day post-irradiation, the number of micronuclei reached 1403 occasions as compared with 1787 occasions on the first day post-irradiation. (Table 10) .

The percentage of total micronuclei after the third day of irradiation reached nearly 23.38% . The bone marrow activity reach 0.023383 after third day post-irradiation (Fig, 18).

After the seventh day post-irradiation, the micronuclei reached 1031 occasions as compared with 1787 of the first day and 1403 after the third day post-irradiation. (Table 11).

The percentage of total micronuclei aberrations after the seventh day post irradiation reached nearly 17.18%. The bone marrow activity reach 0.01718 after seventh day post-irradiation (Table 11 & Fig, 19).

After the fifteen and thirty day post-irradiation, the total micronuclei reached 265 and 255 respectively and the percentage of total micronuclei after fifteen and thirty day post- irradiation reached 4.4% and 4.25% respectively and the bone marrow activity reach 0.04416 and 0.0425 after fifteen and thirty day post-irradiation (Table 12 & 13) and (Fig, 20, 21). Moreover the comparison between the percentage of total micronuclei reached 29.78% , 23.38%, 17.18%, 4.4% and 4.25% after first, third, seventh, fifteen and thirty day post-irradiation respectively.

The frequency of micronuclei from rat bone marrow cells was obtained after first, third and seventh day post-irradiation. Very highly significant (Table 14). While after fifteen and thirty day post-irradiation,

there was non significant due to radiation as it was nearly the same as in control groups.

The most harvested MN were round while almond and linear were rare (Fig, 22). Most erythrocytes bi nucleated and multinucleated.

Table (8): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats in control group.

No. of rats	**No. of examined Cells 1000/rat**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	36	4	1	41
R2	1000	41	3	0	44
R3	1000	35	1	0	36
R4	1000	39	2	0	41
R5	1000	44	2	0	46
R6	1000	34	3	1	38
Total	6000	229	15	2	246
Mean	1000	38.16	2.5	0.33	41.83
%		3.81	0.25%	0.03	4.1
S.E.		1.57	0.42	0.21	1.50

Table (9): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of rats irradiated with 6 Gy after one day.

No. of rats	**No. of examined Cells 1000/rat**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	311	23	8	342
R2	1000	256	30	5	291
R3	1000	260	25	6	291
R4	1000	247	31	5	283
R5	1000	276	25	8	309
R6	1000	236	28	7	271
Total	6000	1586	162	39	1787
Mean	1000	264.33	27	6.5	297.83
%		26.43	2.7	0.65	29.78
S.E.		10.80	1.29	0.56	10.17

Table (10): frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after third day.

No. of rats	**No. of examined Cells 1000/rat**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	202	27	6	235
R2	1000	205	8	4	217
R3	1000	212	14	6	232
R4	1000	200	18	10	228
R5	1000	218	12	8	238
R6	1000	222	20	11	253
Total	6000	1259	99	45	1403
Mean	1000	209.83	16.5	7.5	233.83
%		20.98	1.65	0.75	23.38
S.E.		3.65	2.72	1.08	4.85

Table (11): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after seventh day.

No. of rats	**No. of examined Cells 1000/rat**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	135	11	4	150
R2	1000	138	11	3	152
R3	1000	154	12	6	172
R4	1000	167	7	6	180
R5	1000	177	9	5	191
R6	1000	174	8	4	186
Total	6000	945	58	28	1031
Mean	1000	157.5	9.66	4.66	171.83
%		15.75	0.96	0.46	17.18
S.E.		7.39	0.80	0.49	7.08

Table (12): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after fifteen day.

No. of rats	**No. of examined Cells 1000/rat**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	43	3	0	46
R2	1000	44	2	0	46
R3	1000	33	5	0	38
R4	1000	38	1	1	40
R5	1000	41	1	0	42
R6	1000	49	2	2	53
Total	6000	248	14	3	265
Mean	1000	41.33	2.33	0.5	44.16
%		4.13	0.23	0.05	4.4
S.E.		2.23	0.61	0.34	2.19

Table (13): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after 30 day.

No. of rats	**No. of examined Cells 1000/rat**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	45	8	0	53
R2	1000	40	4	0	44
R3	1000	40	3	0	43
R4	1000	43	3	0	46
R5	1000	34	1	0	35
R6	1000	32	2	0	34
Total	6000	234	21	0	255
Mean	1000	39	3.5	0	42.5
%		3.9	0.35	0	4.25
S.E.		2.06	0.99	0.0	2.90

Table (8): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats in control group.

No. of rats	**No. of examind Cells**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	36	4	1	41
R2	1000	41	3	0	44
R3	1000	35	1	0	36
R4	1000	39	2	0	41
R5	1000	44	2	0	46
R6	1000	34	3	1	38
Total	6000	229	15	2	246
Mean	1000	38.16	2.5	0.33	41.83
%		3.81	0.25%	0.03	4.1
S.E.		1.57	0.42	0.21	1.50

Table (9): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of rats irradiated with 6 Gy after one day.

No. of rats	**No. of examind Cells**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	311	23	8	342
R2	1000	256	30	5	291
R3	1000	260	25	6	291
R4	1000	247	31	5	283
R5	1000	276	25	8	309
R6	1000	236	28	7	271
Total	6000	1586	162	39	1787
Mean	1000	264.33	27	6.5	297.83
%		26.43	2.7	0.65	29.78
S.E.		10.80	1.29	0.56	10.17

Table (10): frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after third day.

No. of rats	**No. of examind Cells**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	202	27	6	235
R2	1000	205	8	4	217
R3	1000	212	14	6	232
R4	1000	200	18	10	228
R5	1000	218	12	8	238
R6	1000	222	20	11	253
Total	6000	1259	99	45	1403
Mean	1000	209.83	16.5	7.5	233.83
%		20.98	1.65	0.75	23.38
S.E.		3.65	2.72	1.08	4.85

Table (11): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after seventh day.

No. of rats	**No. of examind Cells**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	135	11	4	150
R2	1000	138	11	3	152
R3	1000	154	12	6	172
R4	1000	167	7	6	180
R5	1000	177	9	5	191
R6	1000	174	8	4	186
Total	6000	945	58	28	1031
Mean	1000	157.5	9.66	4.66	171.83
%		15.75	0.96	0.46	17.18
S.E.		7.39	0.80	0.49	7.08

Table (12): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after fifteen day.

No. of rats	**No. of examind Cells**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	43	3	0	46
R2	1000	44	2	0	46
R3	1000	33	5	0	38
R4	1000	38	1	1	40
R5	1000	41	1	0	42
R6	1000	49	2	2	53
Total	6000	248	14	3	265
Mean	1000	41.33	2.33	0.5	44.16
%		4.13	0.23	0.05	4.4
S.E.		2.23	0.61	0.34	2.19

Table (13): Frequencies of micronuclei polychromatic erythrocyte cells (MNPCEs) from bone marrow cells of male rats irradiated with 6 Gy after 30 day.

No. of rats	**No. of examind Cells**	(MNPCEs) **+ ve**			**Total** (MNPCEs) **+ ve**
		One	**Two**	**Multi**	
R1	1000	45	8	0	53
R2	1000	40	4	0	44
R3	1000	40	3	0	43
R4	1000	43	3	0	46
R5	1000	34	1	0	35
R6	1000	32	2	0	34
Total	6000	234	21	0	255
Mean	1000	39	3.5	0	42.5
%		3.9%	0.35%	0	4.25%
S.E.		2.06	0.99	0.0	2.90

Table (14): Comparison between the frequencies of Micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rats irradiated with 6Gy gamma radiation.

Treatment	*No. of Rats*	*No. of Scored cells 50/rat*	**Micronuclei polychromatic erythrocytes (MNPCEs)** *Aberrations*			*Bone Marrow Activity*
			Total aberrations			
			No.	*%*	*Mean ± SE*	**Mean/100**
Control	**6**	**1000**	**246**	**4.1**	**41 ± 1.50**	**0.041**
1st day	**6**	**1000**	**1787**	**29.78**	**297.83 ± 10.17*** **	**0.2978**
3rd day	**6**	**1000**	**1403**	**23.38**	**233.83 ± 4.85*** **	**0.2338**
7th day	**6**	**1000**	**1031**	**17.18**	**171.83 ± 7.08*** **	**0.1718**
15th day	**6**	**1000**	**265**	**4.4**	**44.16 ± 2.19**	**0.04416**
30th day	**6**	**1000**	**255**	**4.25**	**42.5 ± 2.90**	**0.0425**

*** Significant at P < 0.05** **** Significant at P < 0.01** ***** Significant at P < 0.001**

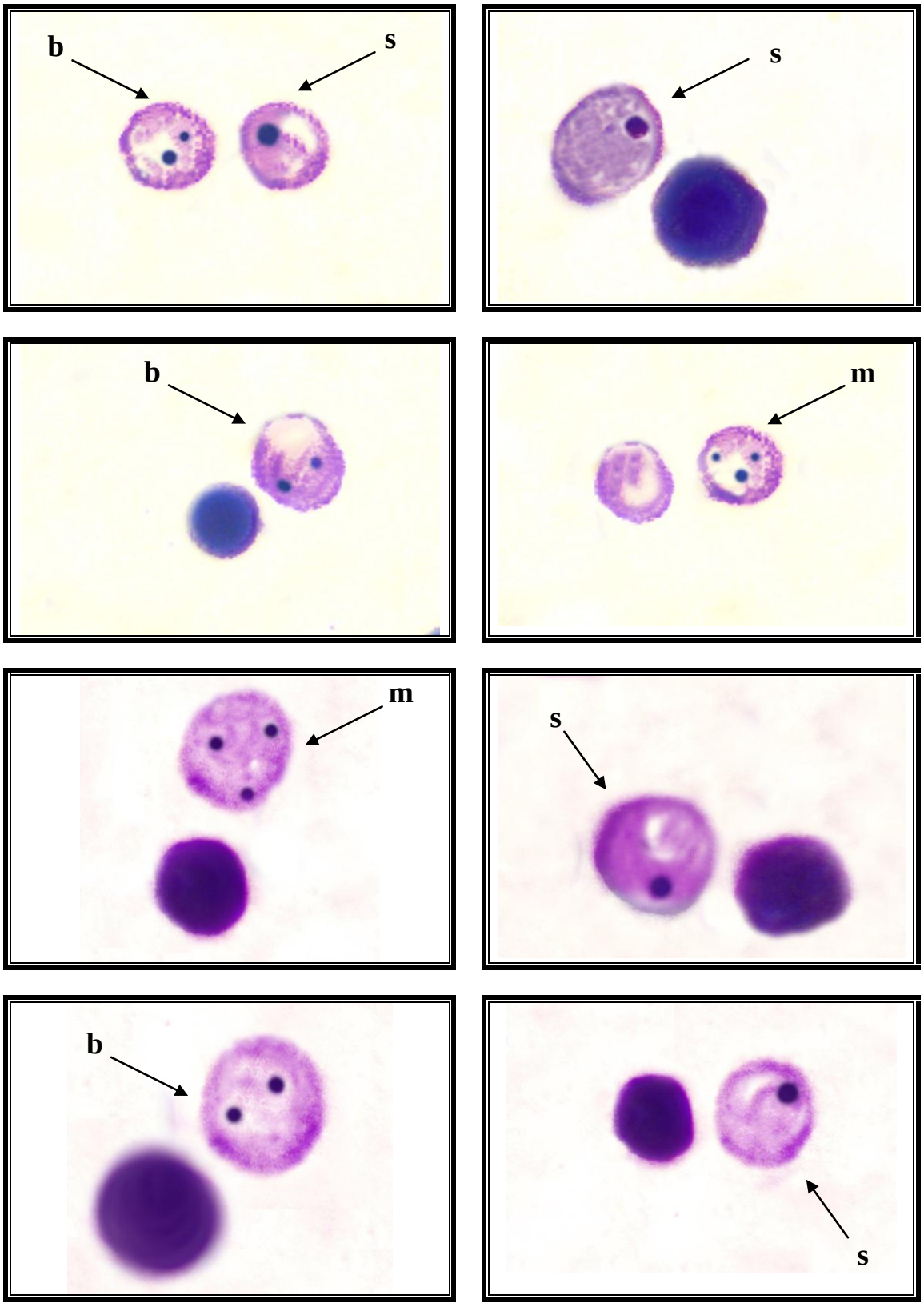


Fig (15): Several micronuclei from bone marrow cells of male rats (single micronuclei), s, (bi micronuclei), b, (multi micronuclei), m.

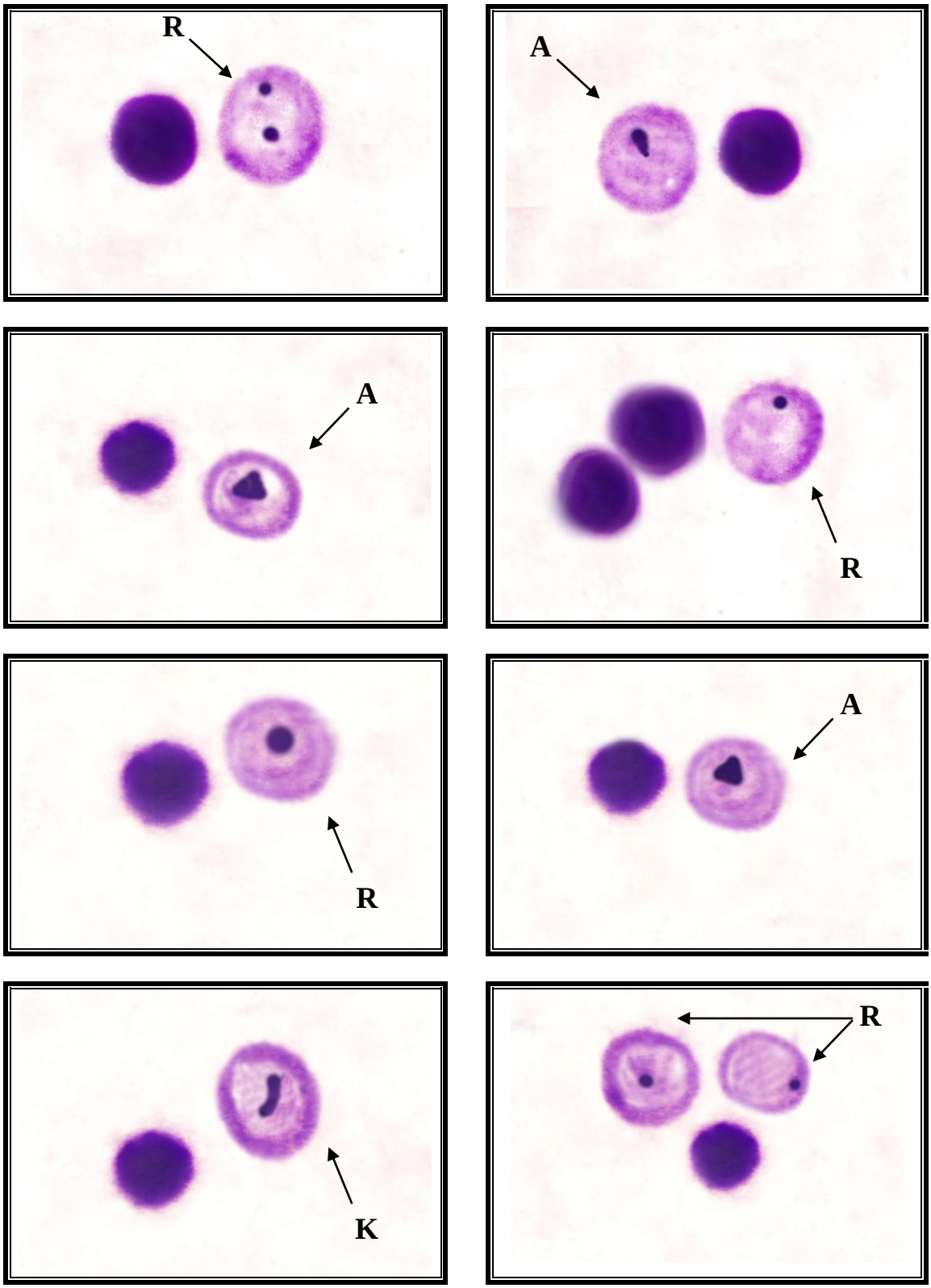


Fig (16): Several micronuclei from bone marrow cells of male rats (Round), R, (Rock), K, (Almond), A.

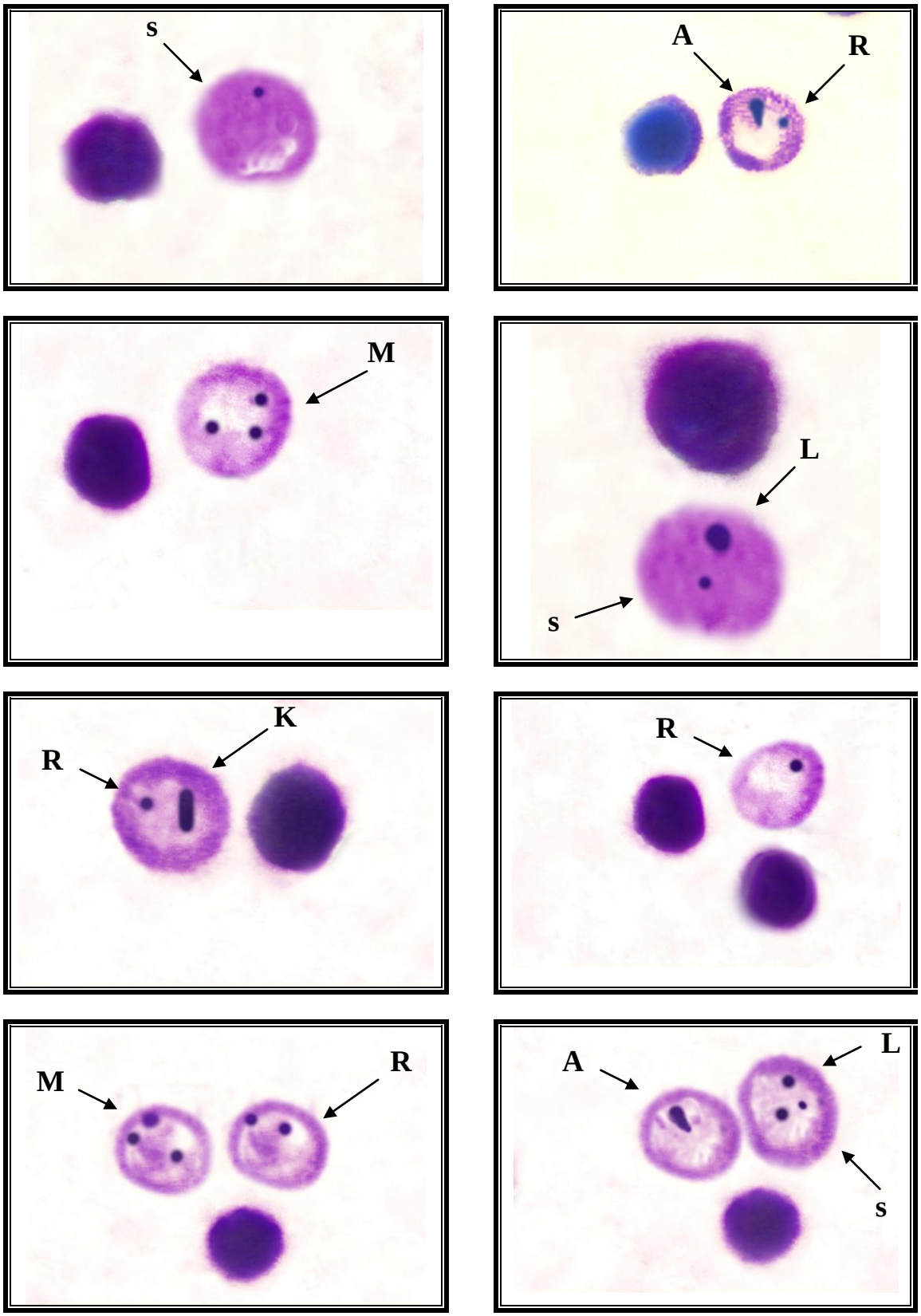


Fig (22): Several erythrocytes from bone marrow cells of irradiated rats with 6Gy containing micronuclei (small MN), s, (large micronuclei), L, (Almond), A, (Rock), K, (Round), R, (Multi micronuclei), M.

Table (14): Comparison between the frequencies of Micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rats irradiated with 6Gy gamma radiation.

Treatment	*No. of Rats*	*No. of examined Cells 1000/rat*	Micronuclei polychromatic erythrocytes (MNPCEs)			*Bone Marrow Activity* Mean/1000
			No.	*%*	*Mean ± SE*	
Control	6	6000	246	4.1	41 ± 1.50	0.041
1st day	6	6000	1787	29.78	297.83 ± 10.17***	0.2979
3rd day	6	6000	1403	23.38	233.83 ± 4.85***	0.2339
7th day	6	6000	1031	17.18	171.83 ± 7.08***	0.1719
15th day	6	6000	265	4.4	44.16 ± 2.19	0.04416
30th day	6	1000	255	4.25	42.5 ± 2.90	0.0425

*** Significant at P < 0.05** **** Significant at P < 0.01** ***** Significant at P < 0.001**

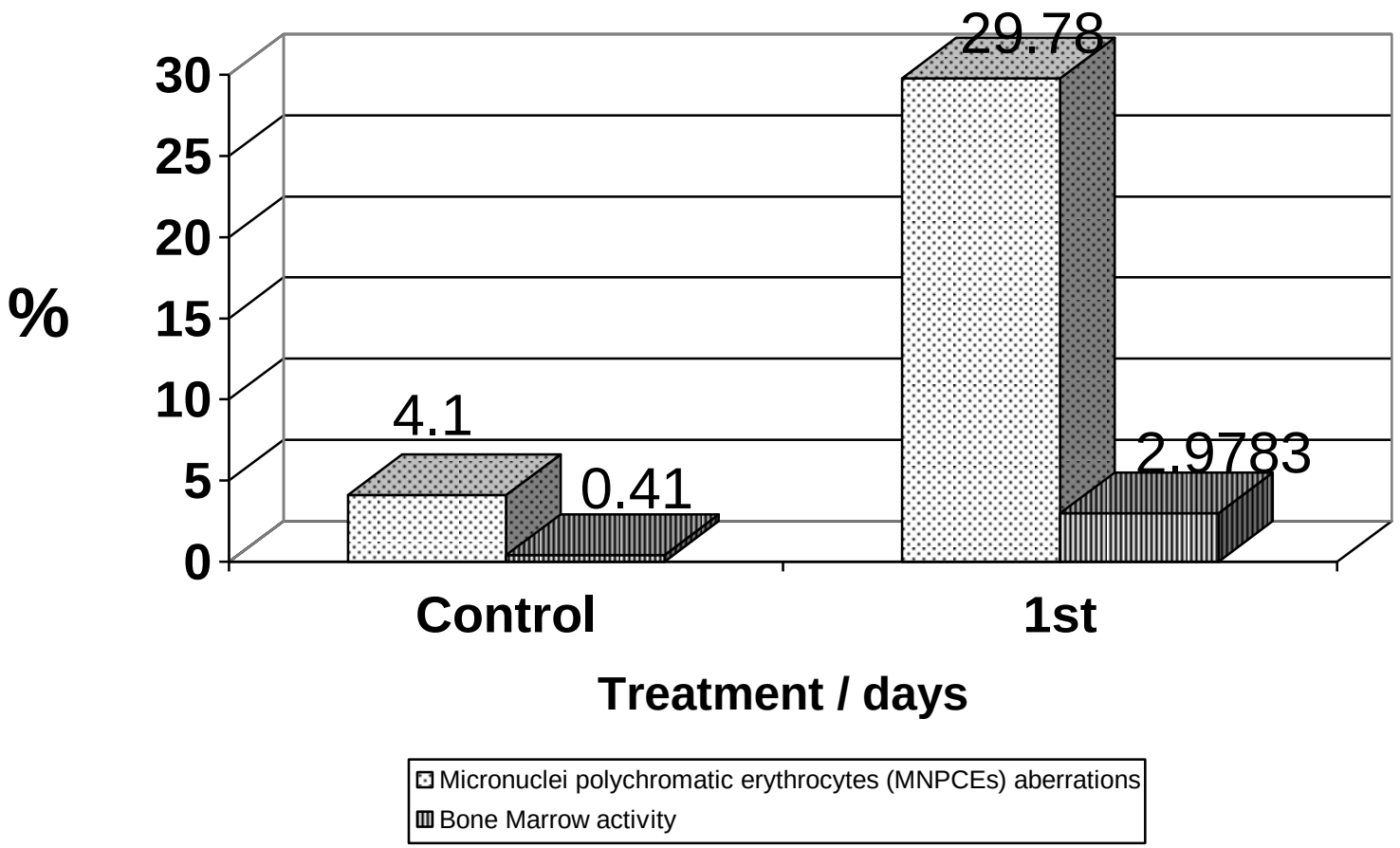


Fig (17): Comparison between the percentage of micronuclei polychromatic erythrocytes (MNPCEs) and bone marrow activity from the bone marrow cells of male rats irradiated with 6Gy gamma irradiation after one day.

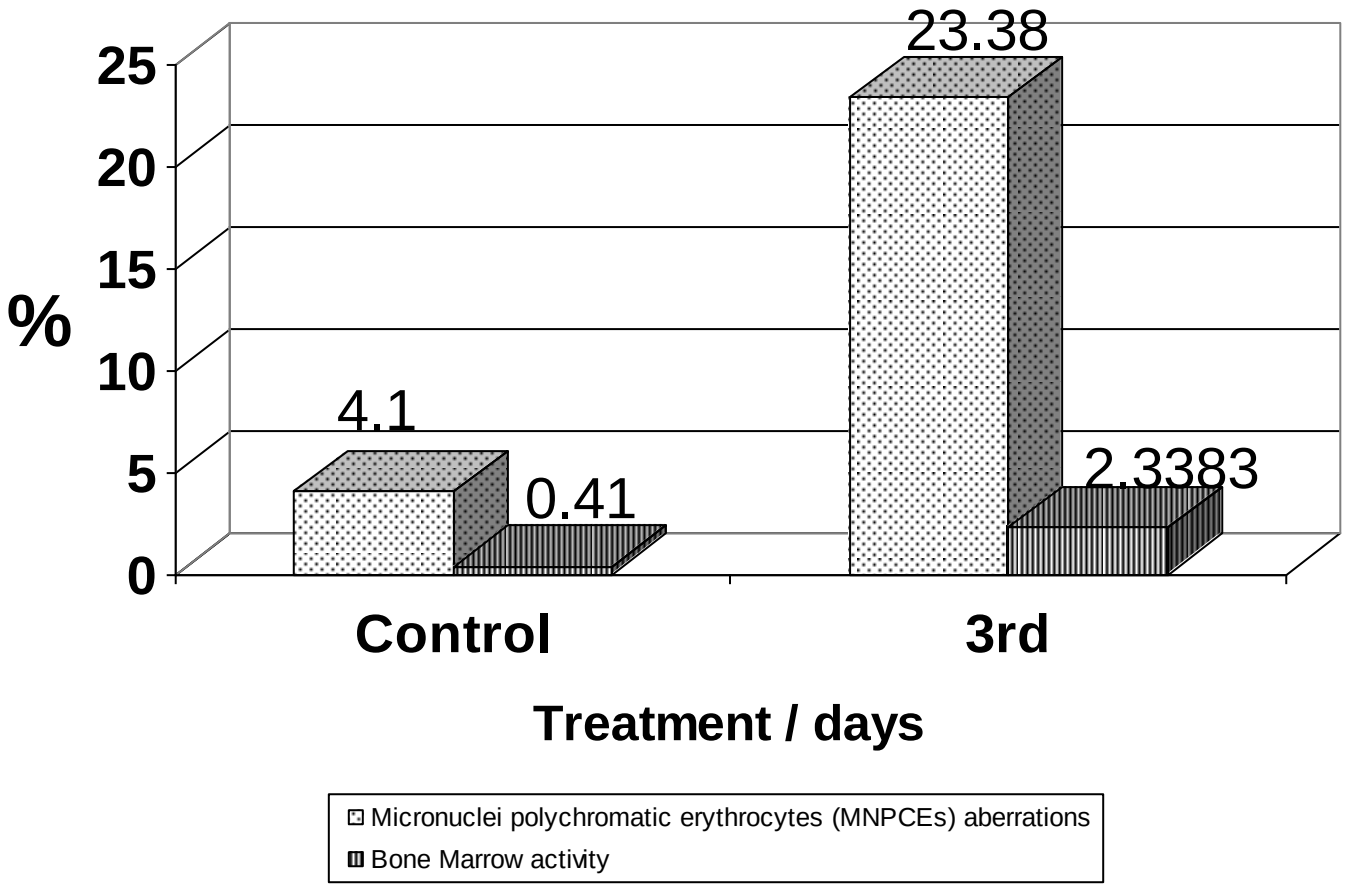


Fig (18): Comparison between the percentage of micronuclei polychromatic erythrocytes (MNPCEs) and bone marrow activity from the bone marrow cells of male rats irradiated with 6Gy gamma irradiation after 3rd day.

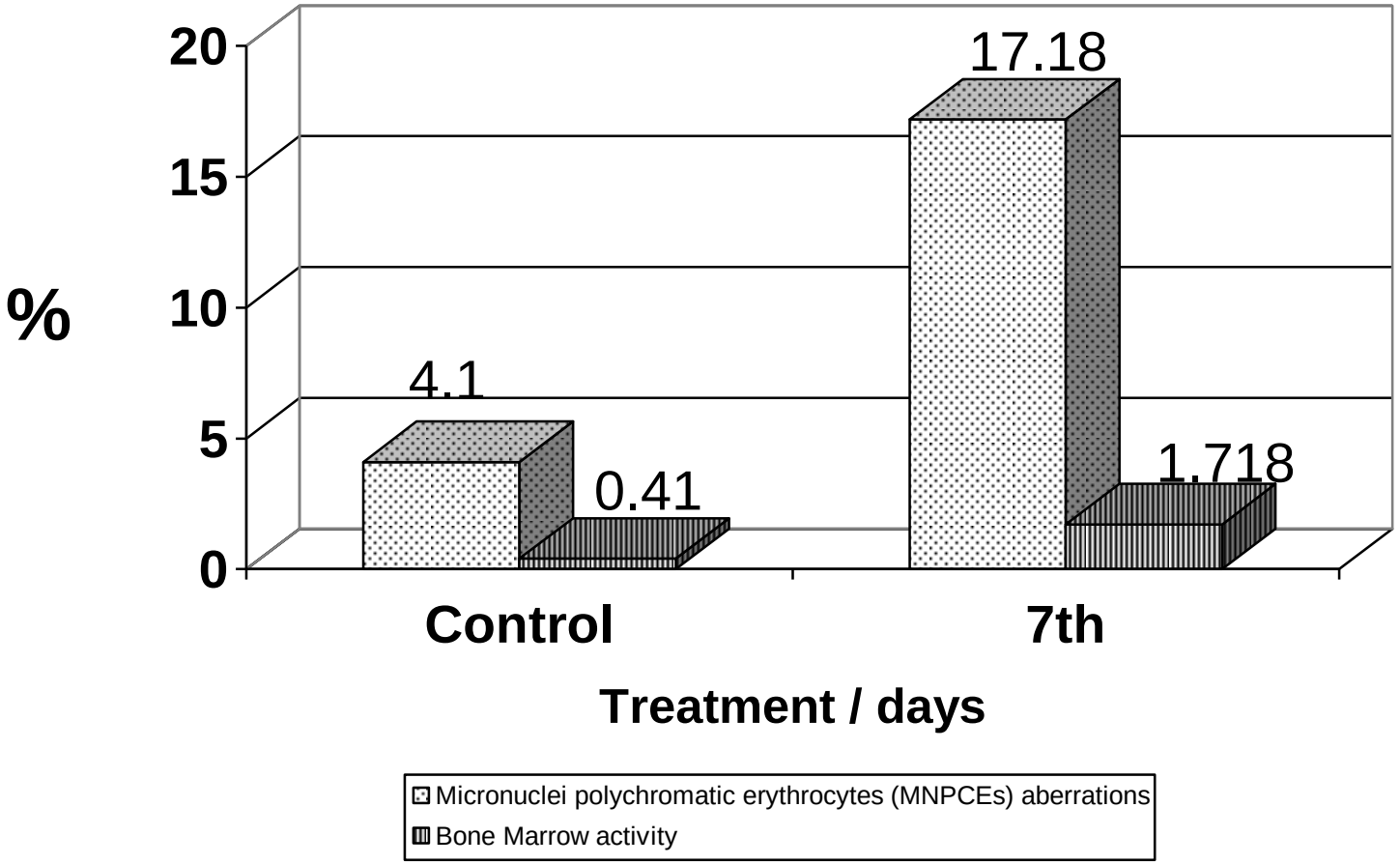


Fig (19): Comparison between the percentage of micronuclei polychromatic erythrocytes (MNPCEs) and bone marrow activity from the bone marrow cells of male rats irradiated with 6Gy gamma irradiation after 7th day.

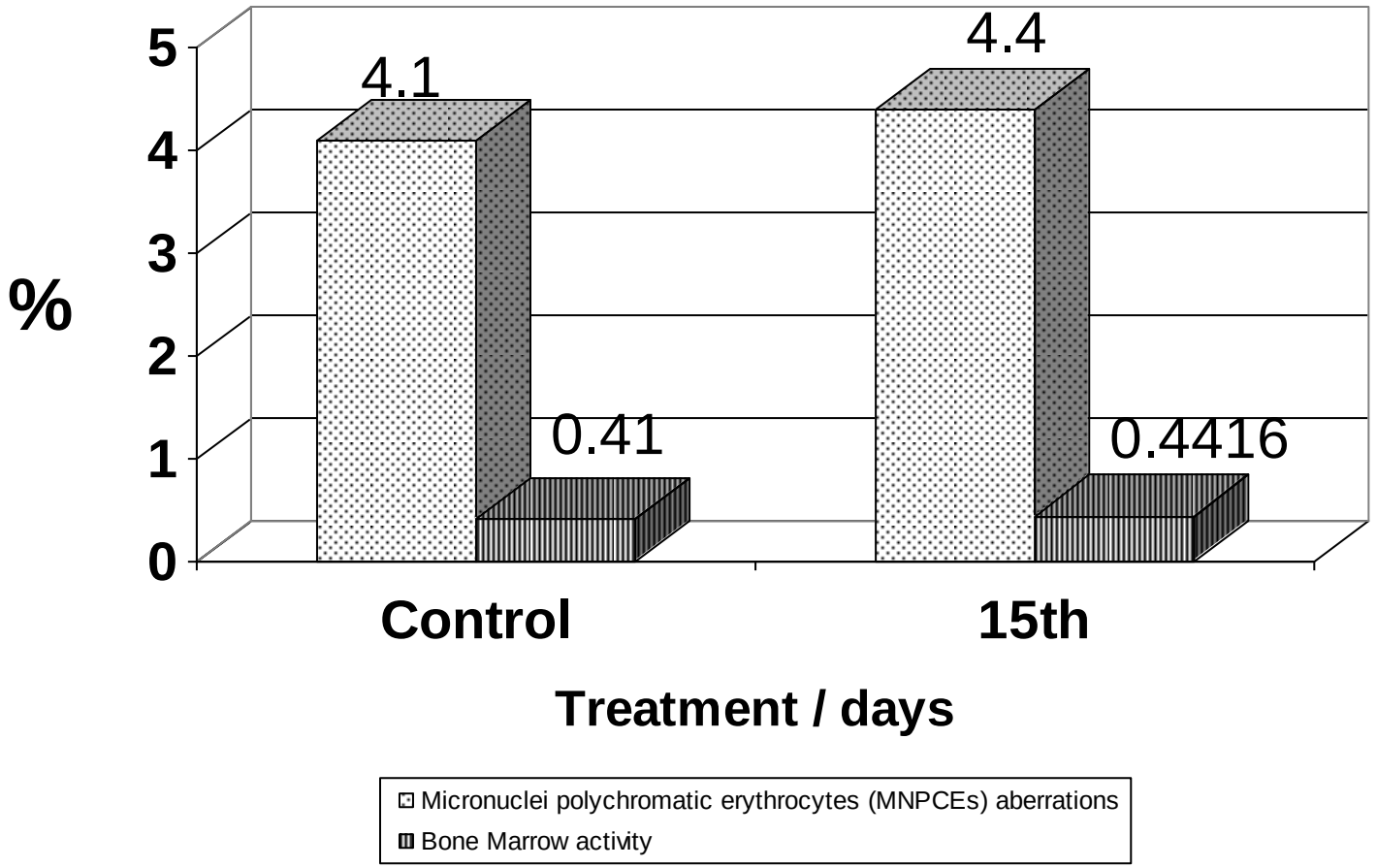


Fig (20): Comparison between the percentage of micronuclei polychromatic erythrocytes (MNPCEs) and bone marrow activity from the bone marrow cells of male rats irradiated with 6Gy gamma irradiation after 15th day.

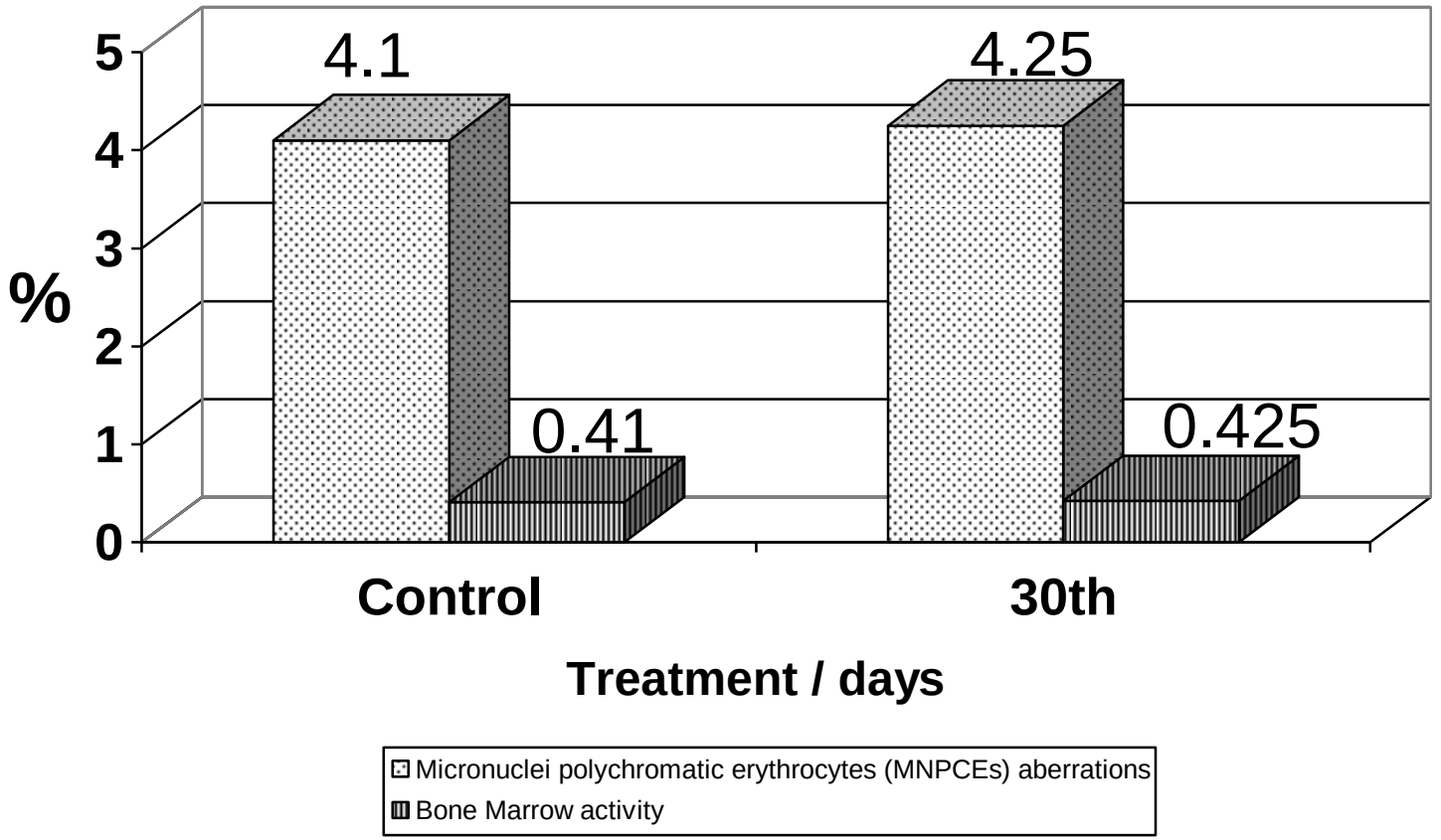


Fig (21): Comparison between the percentage of micronuclei polychromatic erythrocytes (MNPCEs) and bone marrow activity from the bone marrow cells of male rats irradiated with 6Gy gamma irradiation after 30th day.

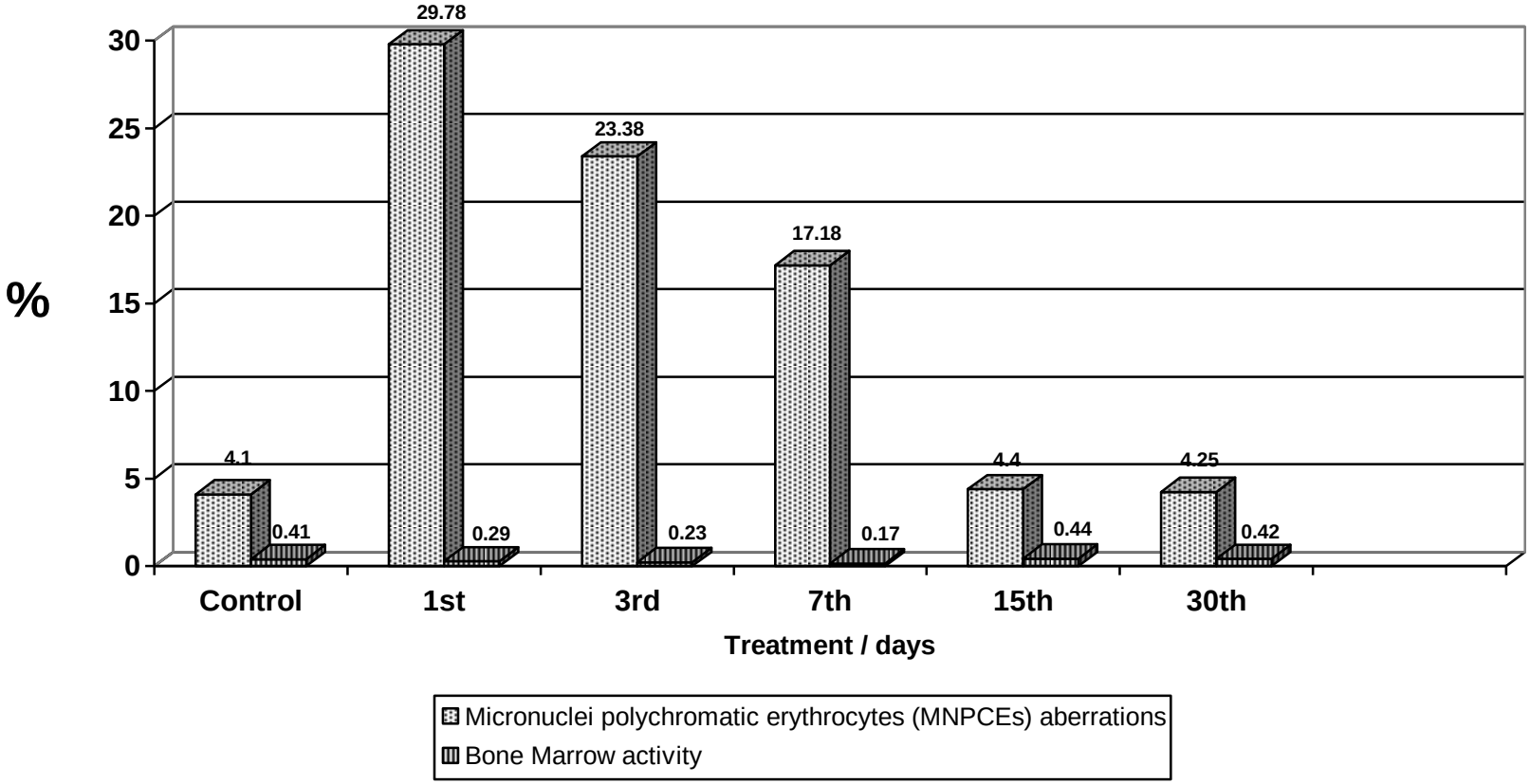


Fig (23): Comparison between the percentage of micronuclei and bone marrow activity from bone marrow cells of male rats irradiated with 6Gy gamma radiation.

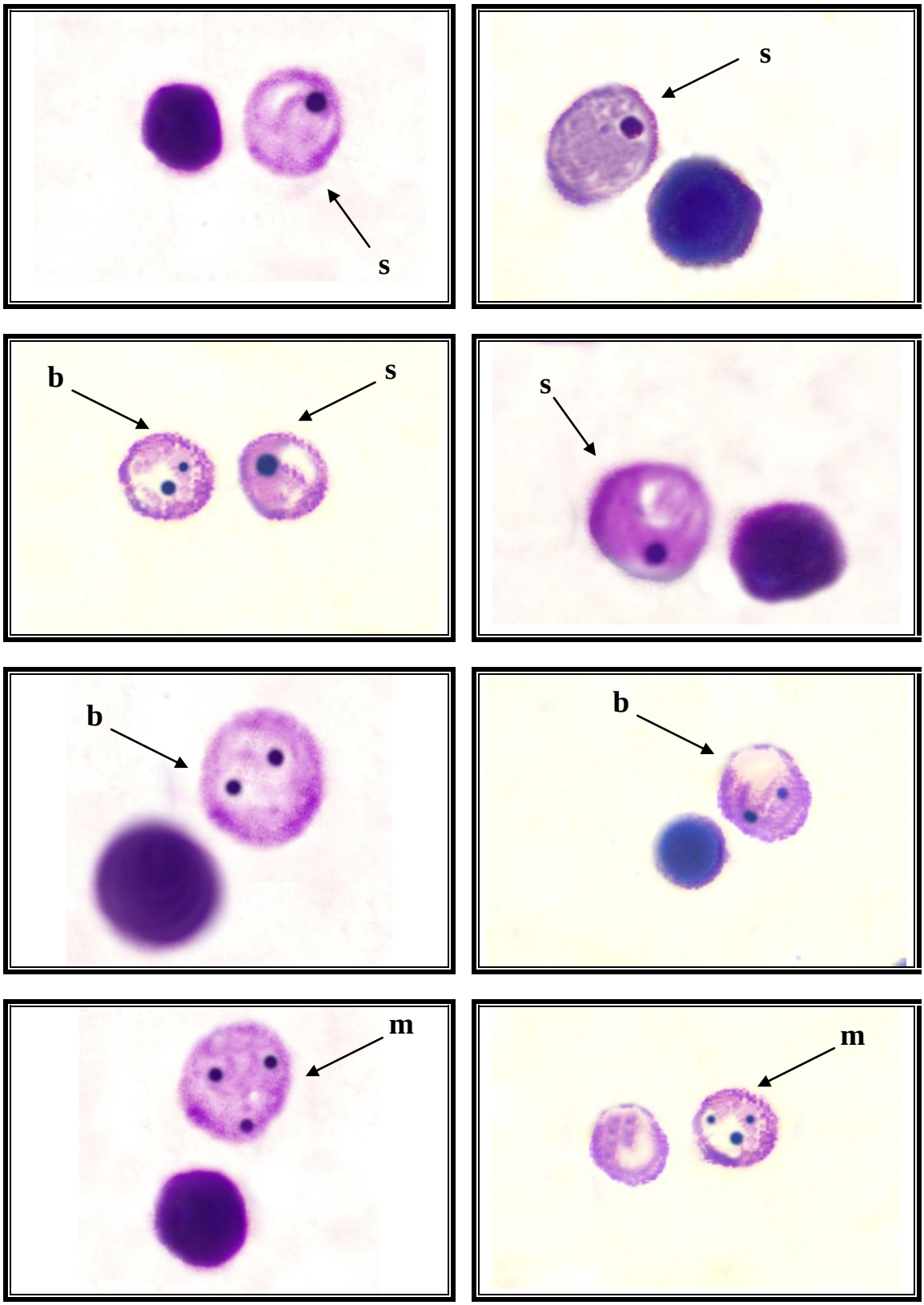


Fig (15): Several MNPCEs from bone marrow cells of male rat single MN (s), bi MN (b), multi MN (m).

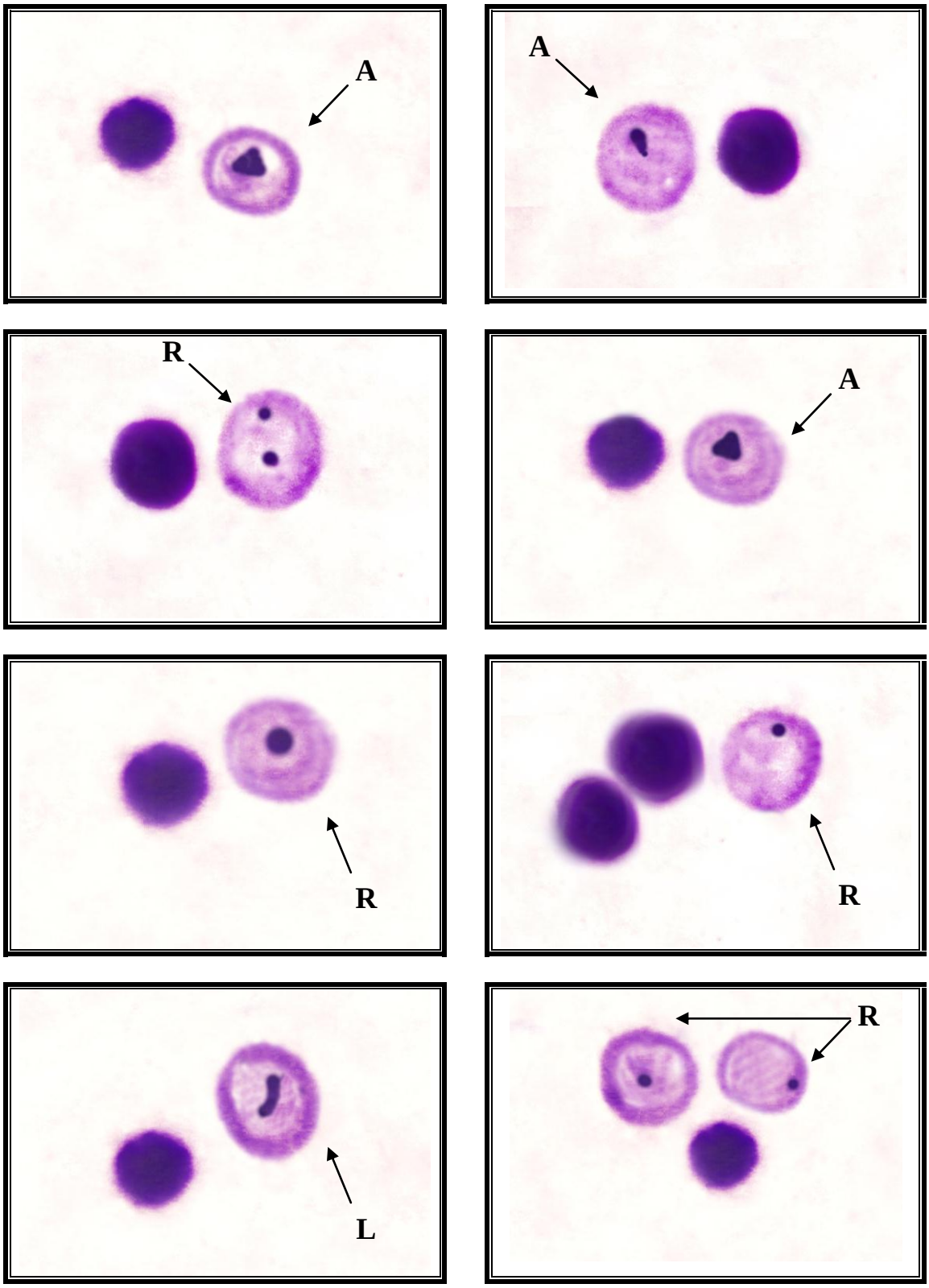


Fig (16): Several MNPCEs with variation shapes from bone marrow cells of irradiated rat Round (R), Linear (L), Almond (A).

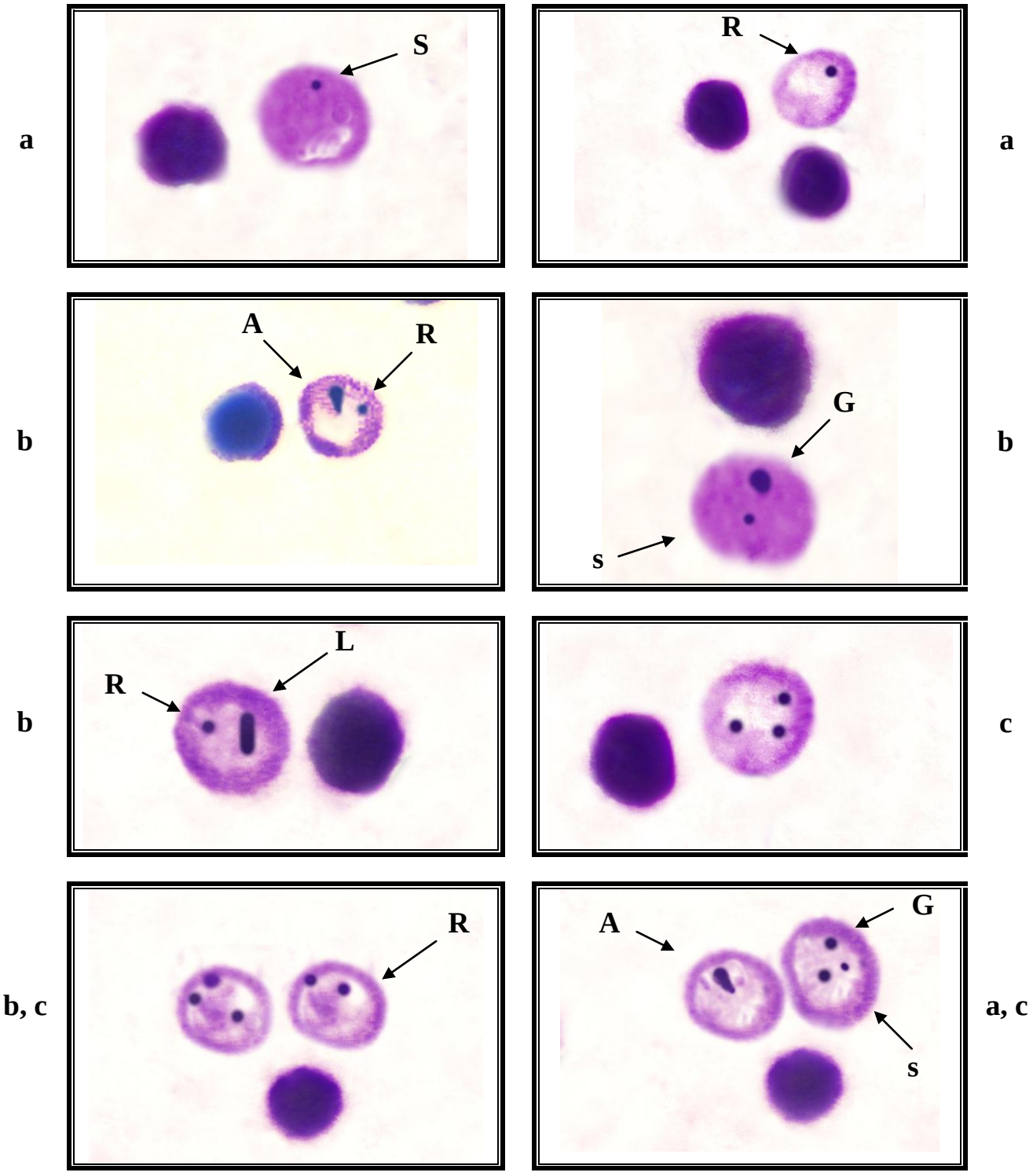


Fig (22): Several polychromatic erythrocytes from bone marrow cells of irradiated rats with 6Gy containing micronuclei with variation size, number and shape Small MN (S), Large MN (G), Almond MN (A), Round MN (R), Linear MN (L).

a- single MN

b- bi MN

c- multi MN

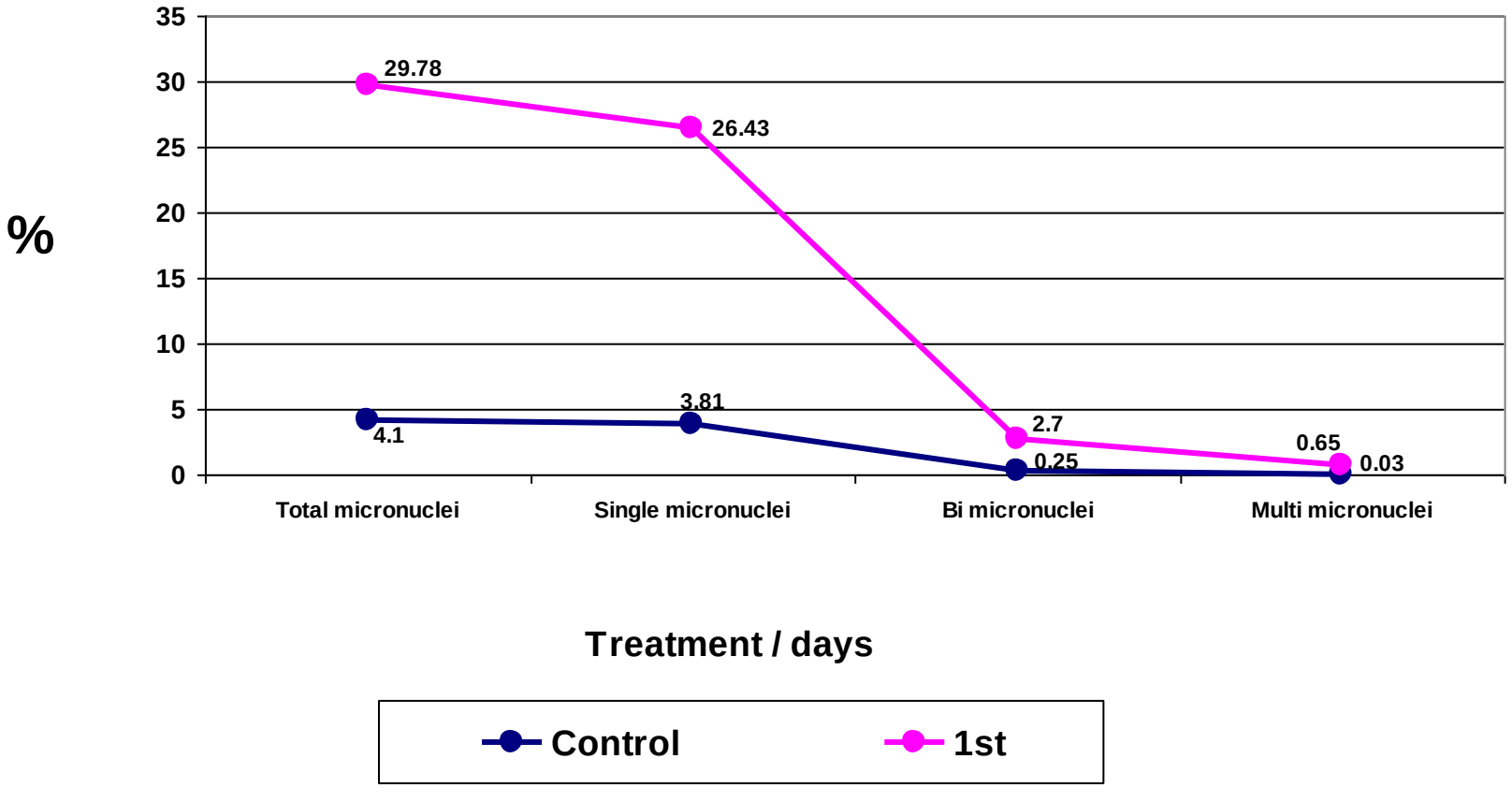


Fig (17): The percentage of micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rat irradiated with 6Gy gamma irradiation after one day.

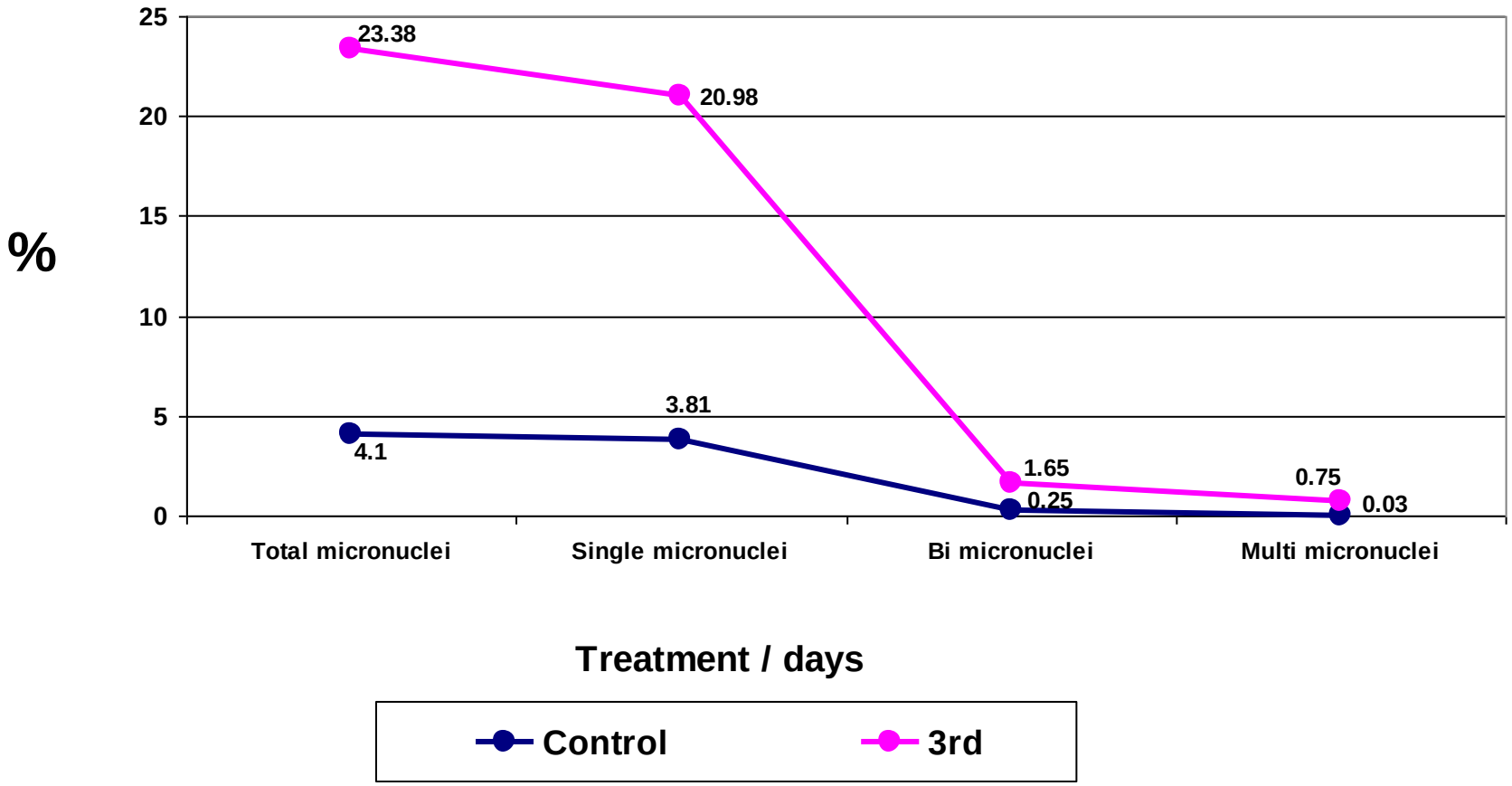


Fig (18): The percentage of micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rat irradiated with 6Gy gamma irradiation after third day.

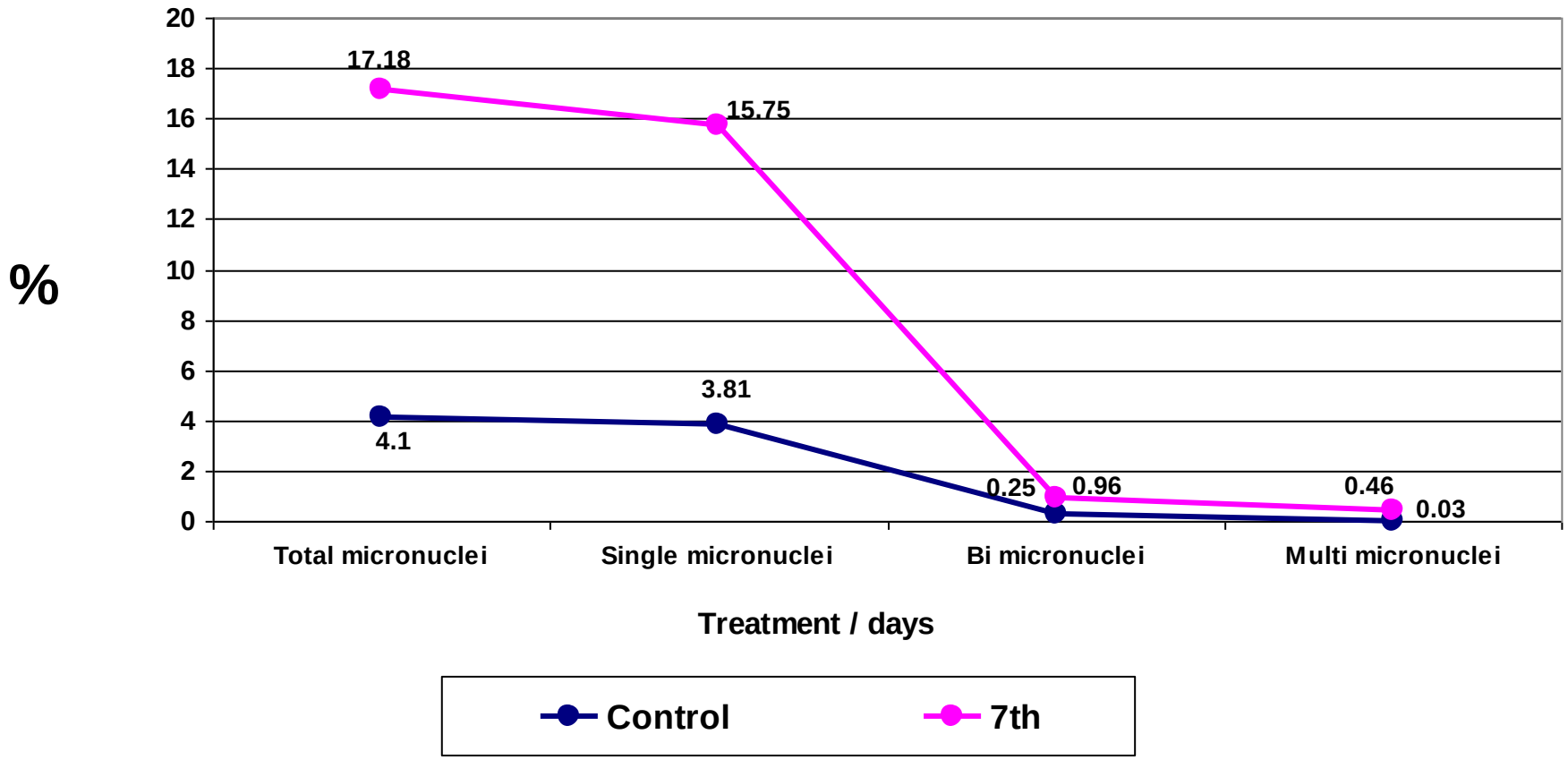


Fig (19): The percentage of micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rat irradiated with 6Gy gamma irradiation after seventh day.

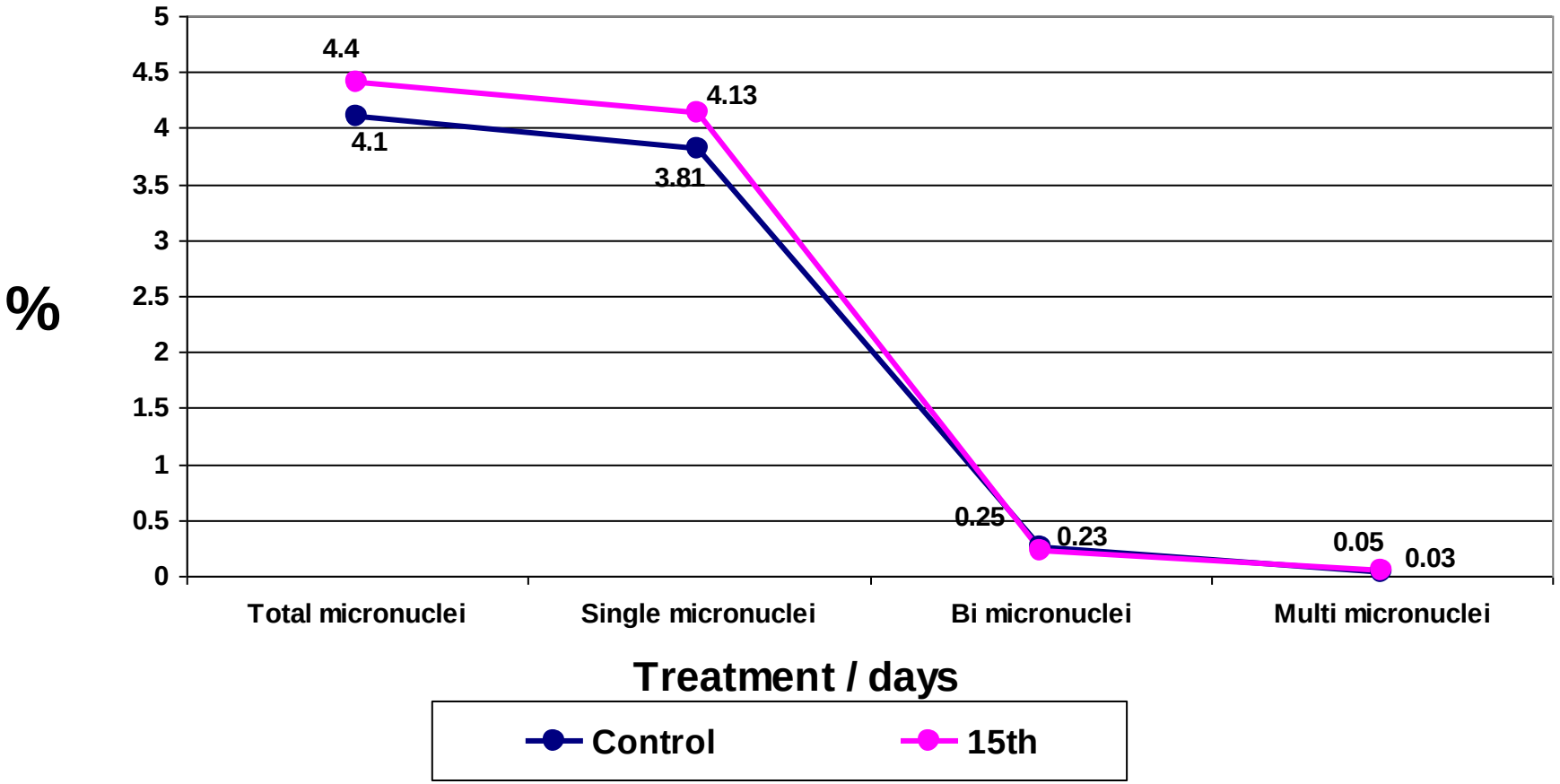


Fig (20): The percentage of micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rat irradiated with 6Gy gamma irradiation after fifth day.

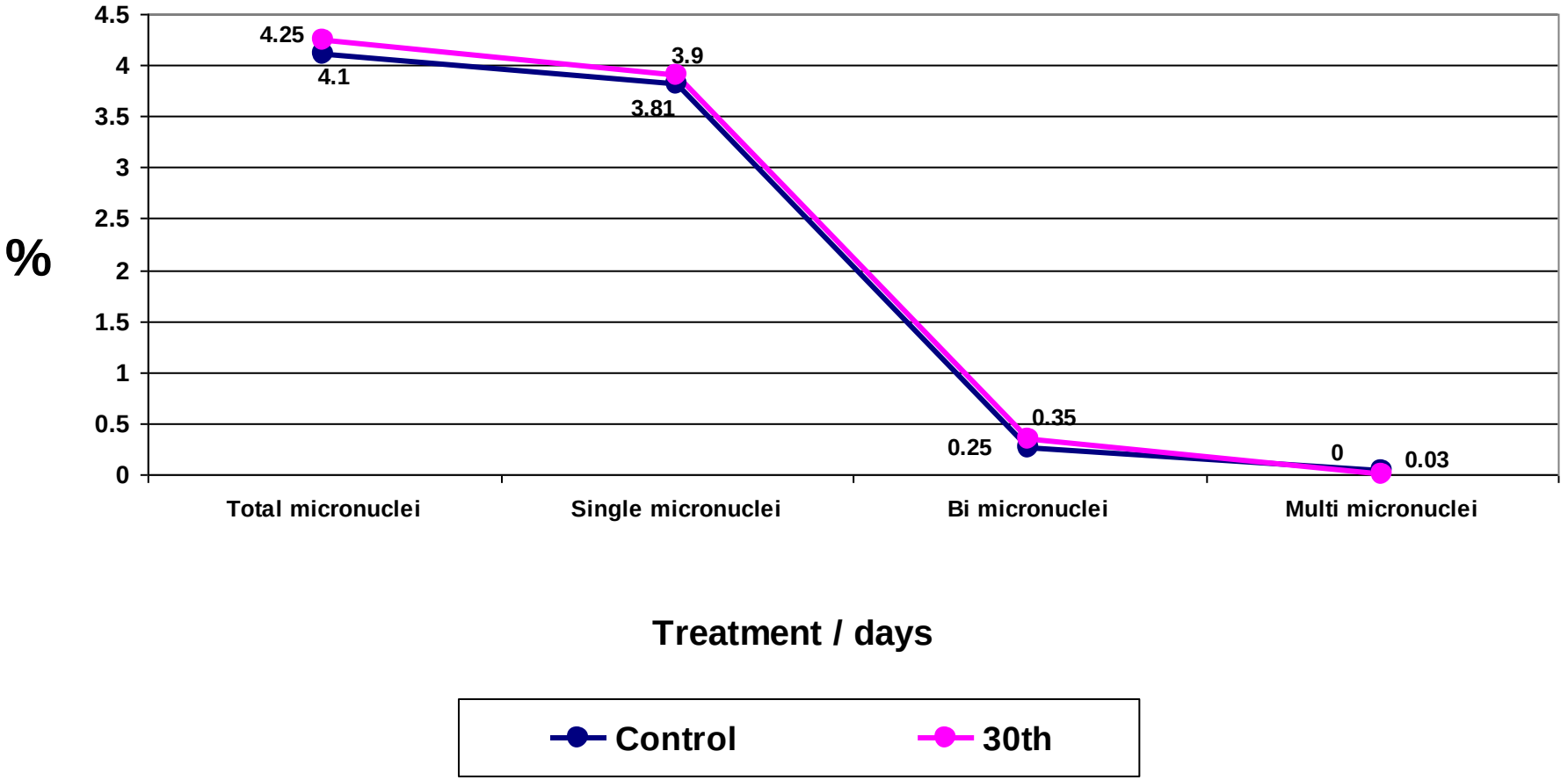


Fig (21): The percentage of micronuclei polychromatic erythrocytes (MNPCEs) from the bone marrow cells of male rat irradiated with 6Gy gamma irradiation after thirty day.

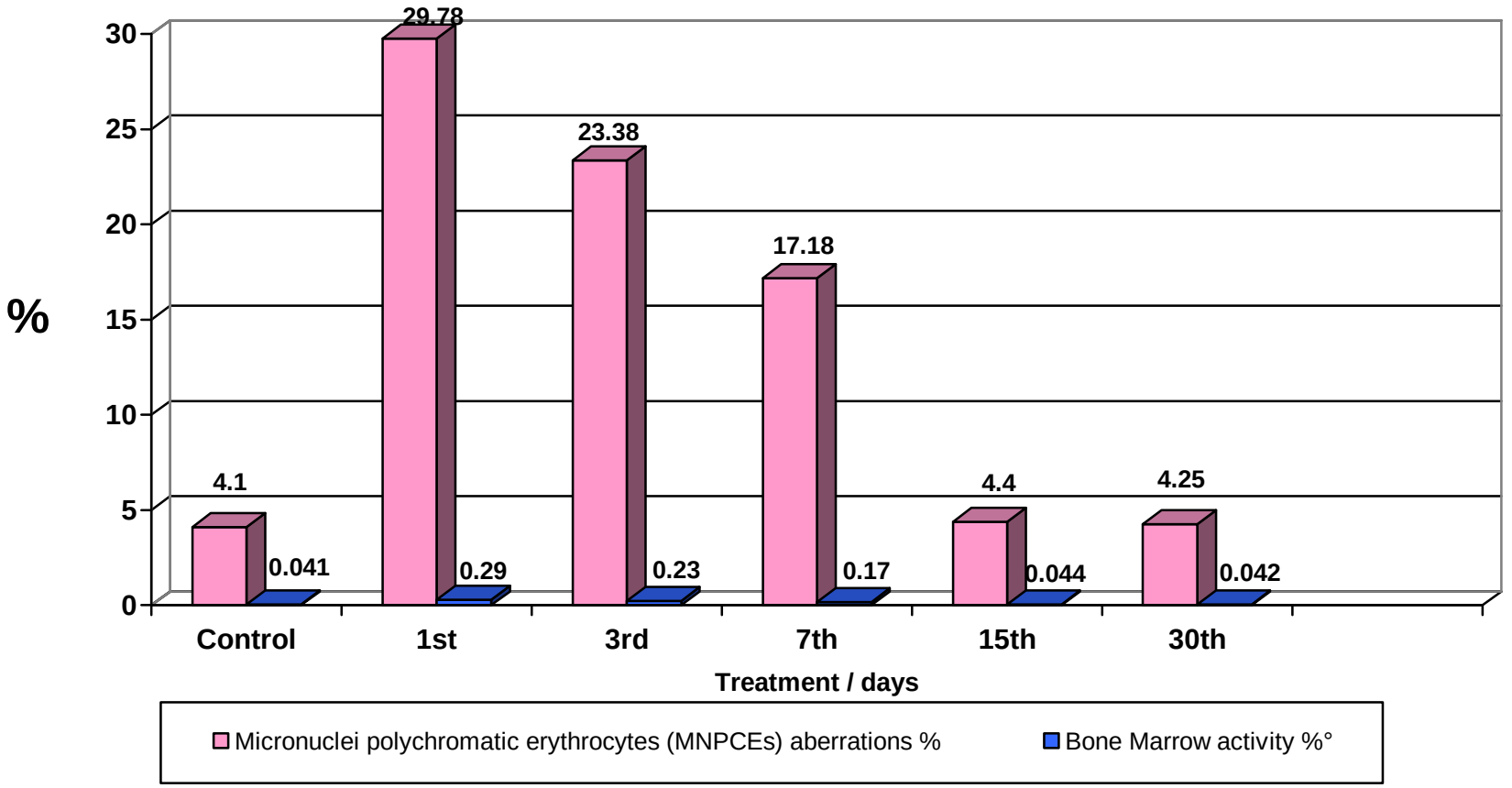


Fig (23): Comparison between the percentage of positive MNPCEs and bone marrow activity of male rat irradiated with 6Gy gamma radiation.

Histochemical observations

General carbohydrates:

A- Normal Rats:

The PAS technique illustrated the carbohydrate material inside the bone marrow cellularity in the form of purple granules distributed in the cytoplasm. Extracellular staining was recognized only in the very thick areas of the film (Fig. 24). Monoblasts appear to contain small amounts of carbohydrate material. The cells of the myelocytic series exihibited a strongly positive reaction, while lymphocytes and those of the erythrocytic series giving little or no reaction respectively.

B- Gamma irradiated rats: -

In rats irradiated at the dose level 6 Gy, the PAS-Positive material in the marrow cells showed a slight decrease in the carbohydrate material in the positive cells after one day post-irradiation as presented in (Fig 25).

On day three post irradiation, a low PAS- Positivity in the reacting cells of the leucocytic and two megakaryocytes as shown in (Fig. 26). This is followed by a persisted decrease in the carbohydrate material inside the positivity regenerating marrow cells which are noticed after seven days post-irradiation as indicated in (Fig. 27).

On day fifteen post-irradiation, the PAS-positivity inside the reacting cells showed a very low degree as indicated in (Fig. 28).

The marrow invironment examined thirty days following irradiation showed a little increase in the PAS positivity inside the reacting cells of regenerating marrow cellularity (Fig. 29).

Total proteins: -

A- Normal rats: -

In the bone marrow cells of adult normal rats, the total proteins appeared in the form of blue color of protein granules characteristic of MBB stain. There was a gradual increase in the intensity of staining in proportion to the stage of maturation of each cell. Lymphoblasts (Lb) exhibited a moderate reaction. The megakaryocytes (Mk) and late normoblasts (LN) showed the highest proteinic content amongst all the marrow cells (Figs 30, 31).

B- Gamma irradiated rats: -

Whole body gamma-irradiation of rats at the exposure dose level of 6Gy, resulted in a slight decrease in the reactivity of the cells inside the damaged marrow environment examined one day post-irradiation (Fig. 32)

On day three post-irradiation, there was a more decrease in the protein content inside the highly destructed nucleated cells as shown in (Fig 33). Except few cells of the polymorphis showed intense reaction for MBB (Fig. 33).

After seven days post-irradiation, there was a high increase in the protein content inside the cells of the leucocytic series and late normoblasts (LN) as shown in (Fig 34).

Partial recovery of the protein content of the marrow cells was detected fifteen days post-irradiation where the marrow cells with variable protein intensities were nearly similar to those of the normal

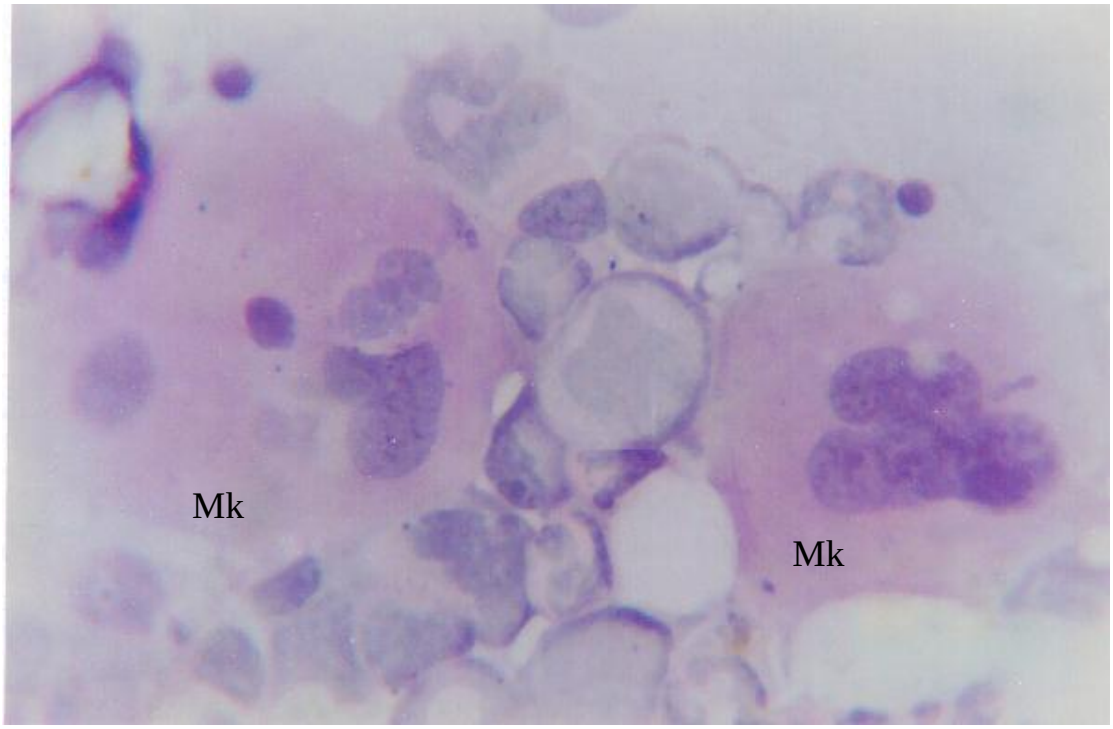
cells. (Fig 35). In the field, two megakaryocytes appeared exihibiting high protein positive materials.

The phase of recovery reached its maximum thirty days post-irradiation, were most cells retained their normal proteinic content in a form nearly similar to that of the normal rats (Fig. 36).

- PAS reaction with no counter stain.

- Fig (24): Bone marrow smear of a normal rat. (PAS) staine showing that the carbohydrate material appeared in the marrow cellularity in the form of purple granules distributed in the cytoplasm. Monoblasts (Mb) appeared to contain small amounts of carbohydrate material. One myelocyte (My) exhibited a strongly +ve PAS reaction. Lymphocytes and those of the erythrocytic series showed a little or no reaction (arrows)

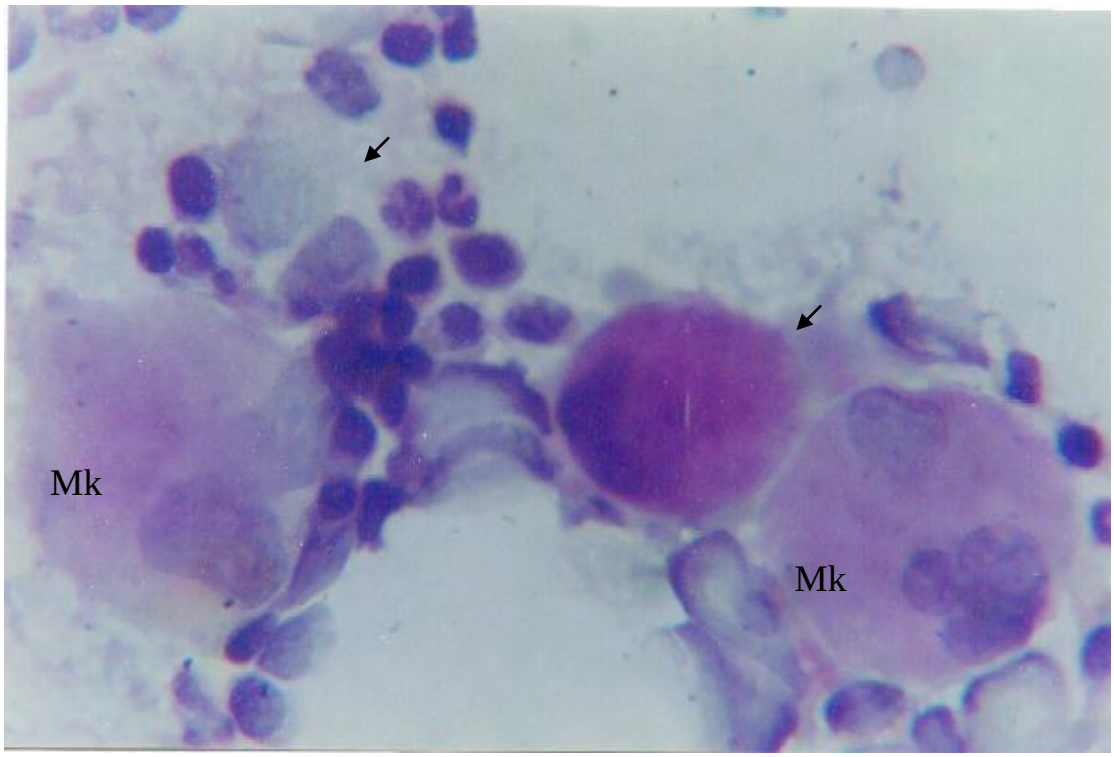
- Original magnification X 1000.



- PAS reaction counter stained with Hx.

- Fig (25): Bone marrow smear of rat, irradiated at 6 Gy gamma rays and examined after one day, showing a slight decrease in the carbohydrate material in the positive cells of the marrow cellularity. In the field two megakaryocytes (Mk) revealed a moderate PAS reaction.

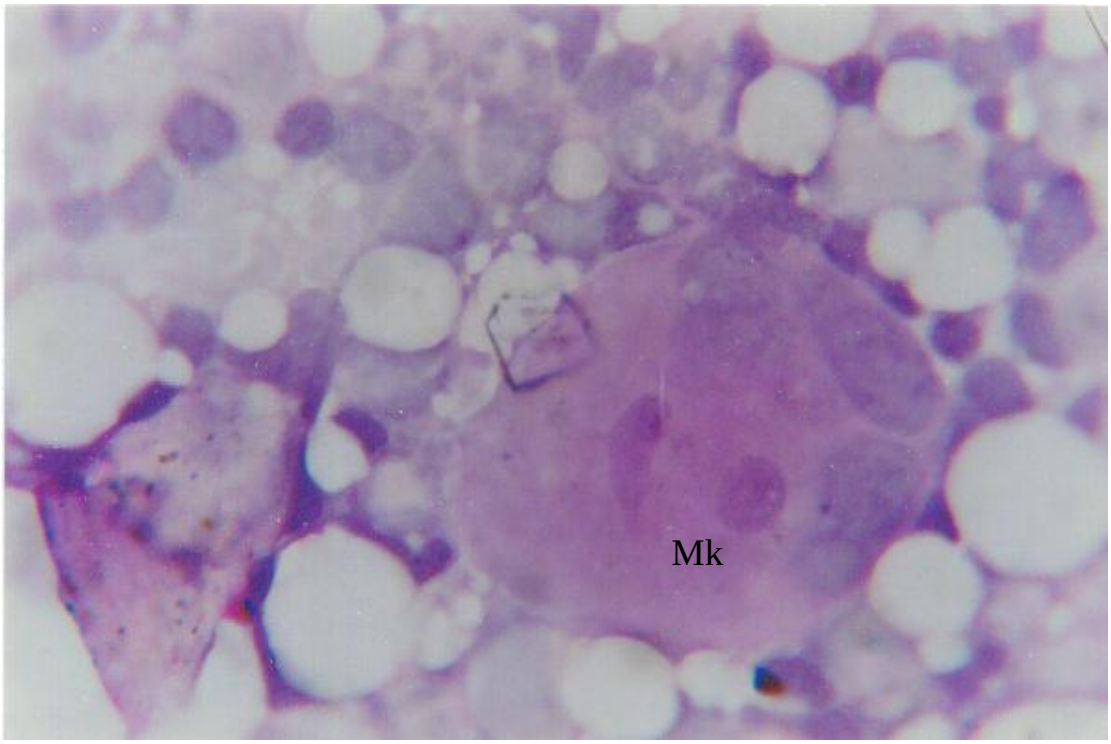
- Original magn. X 1000.



- PAS reaction with Hx counter stain.

- Fig (26): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined three days post-irradiation, showing alone in PAS positivity in the reacting cells of the leucocytic series (arrow) and two megakaryocytes (Mk).

- Original magn. X 1000.

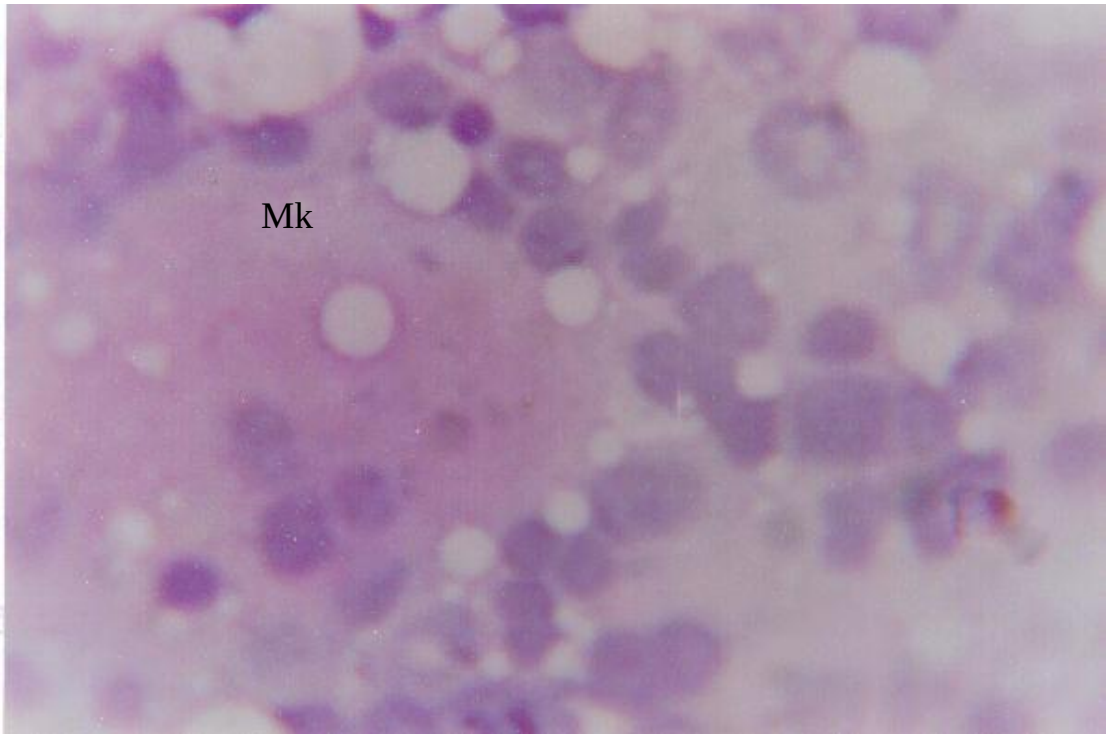


- PAS reaction with Hx counter stain.

- Fig (27): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined seven days post-irradiation, showing persisted low positivity of PAS reaction inside the reacting cells of the partially regenerating marrow environment.

- Original magn. X 1000.

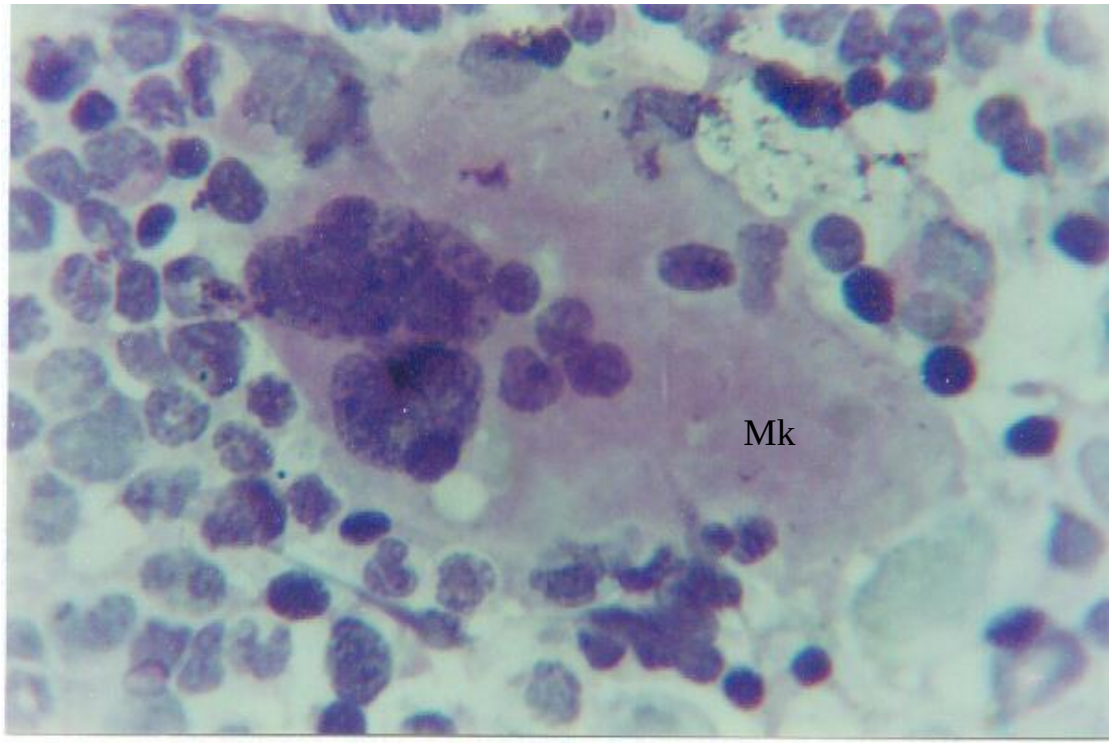
- Megakaryocytes (Mk).



- PAS reaction with Hx counter stain.

- Fig (28): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined fifteen days post-irradiation, showing a very low PAS-positivity inside the reacting cells including a megacaryocyte (Mk).

- Original magn. X 1000.

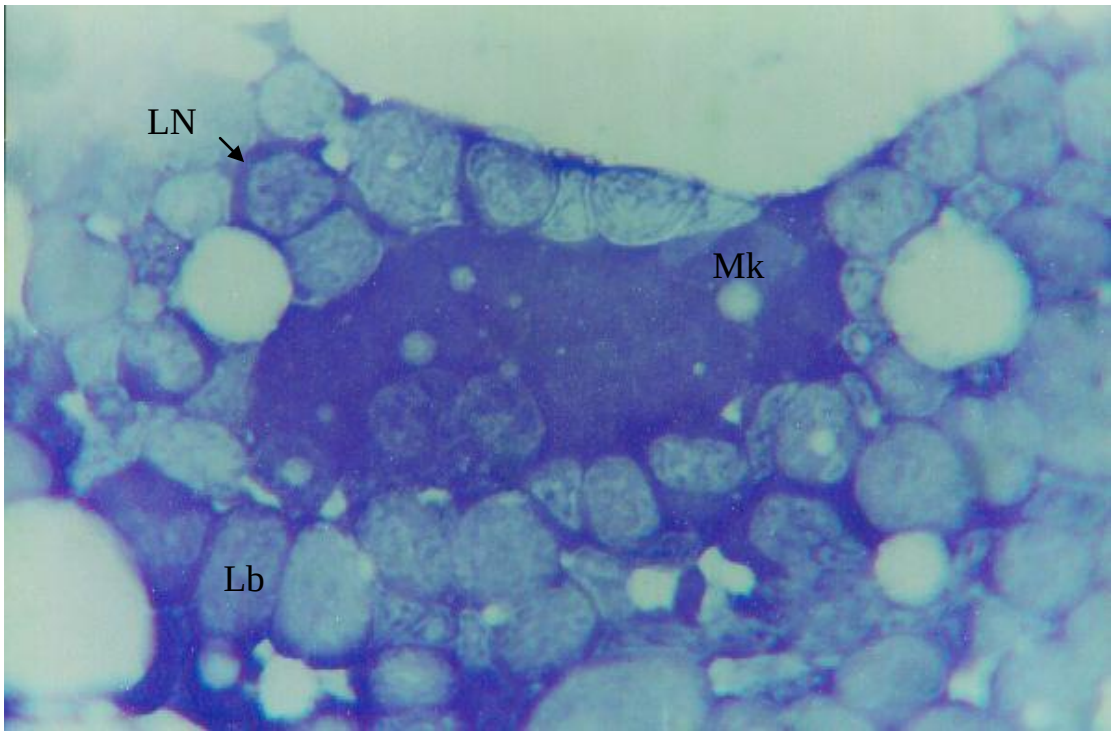


- PAS reaction with Hx counter stain.

- Fig (29): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined thirty days post-irradiation, showing a little increase in PAS-positivity inside the reacting cells of the regenerating marrow cellularity.

- Original magn. X 1000.

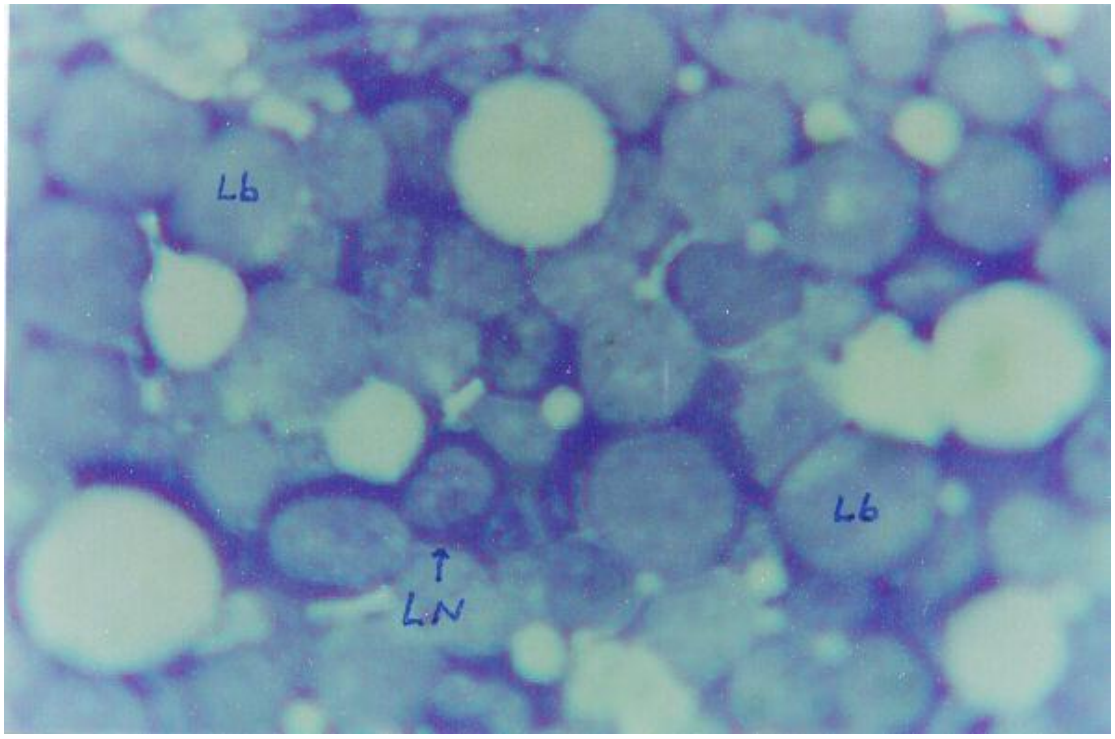
- Megakaryocytes (Mk).



- MBB reaction.

- Fig (30): Bone marrow cells of normal rat showing the characteristic blue colour of the mercury bromophenol blue (MBB) stain inside the marrow cells. Megakaryocytes (Mk) and late normoblast exhibiting the more stronger activity. Lymphoblasts (Lb) showed a moderate reaction.

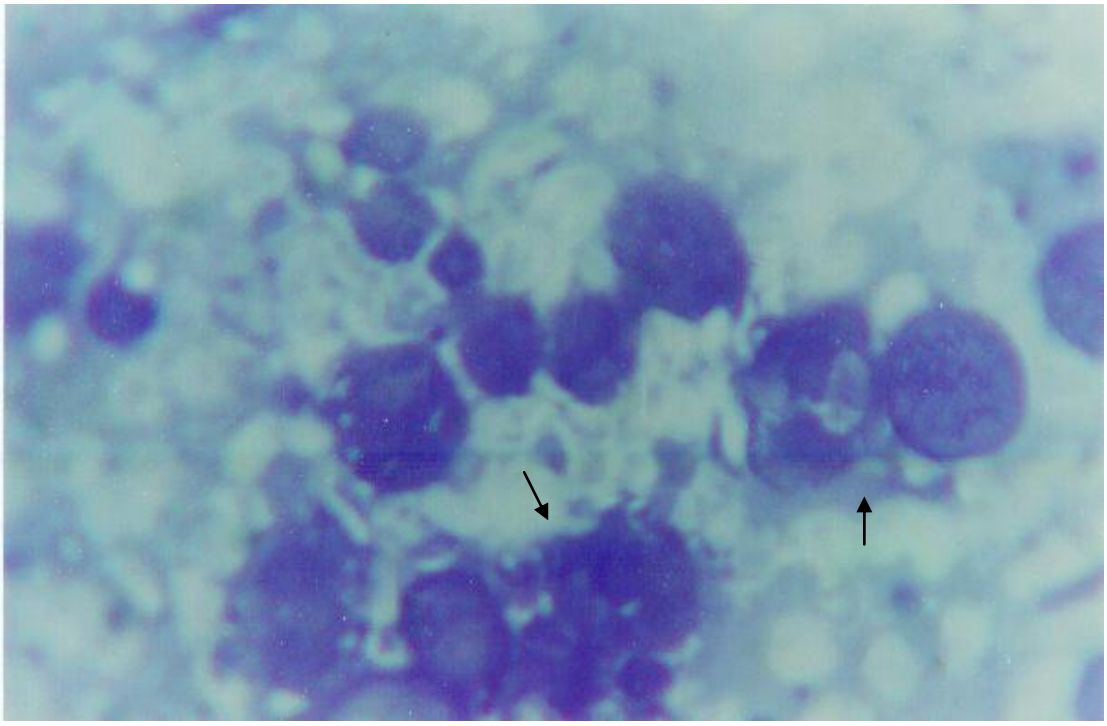
- Original magnification X 1000.



- MBB reaction.

- Fig. (31): Bone marrow cells of normal rats, showing the blue colour of protein granules characteristic of MBB stain inside the marrow cells. Lymphoblasts (Lb). the late normoblast cells (LN).

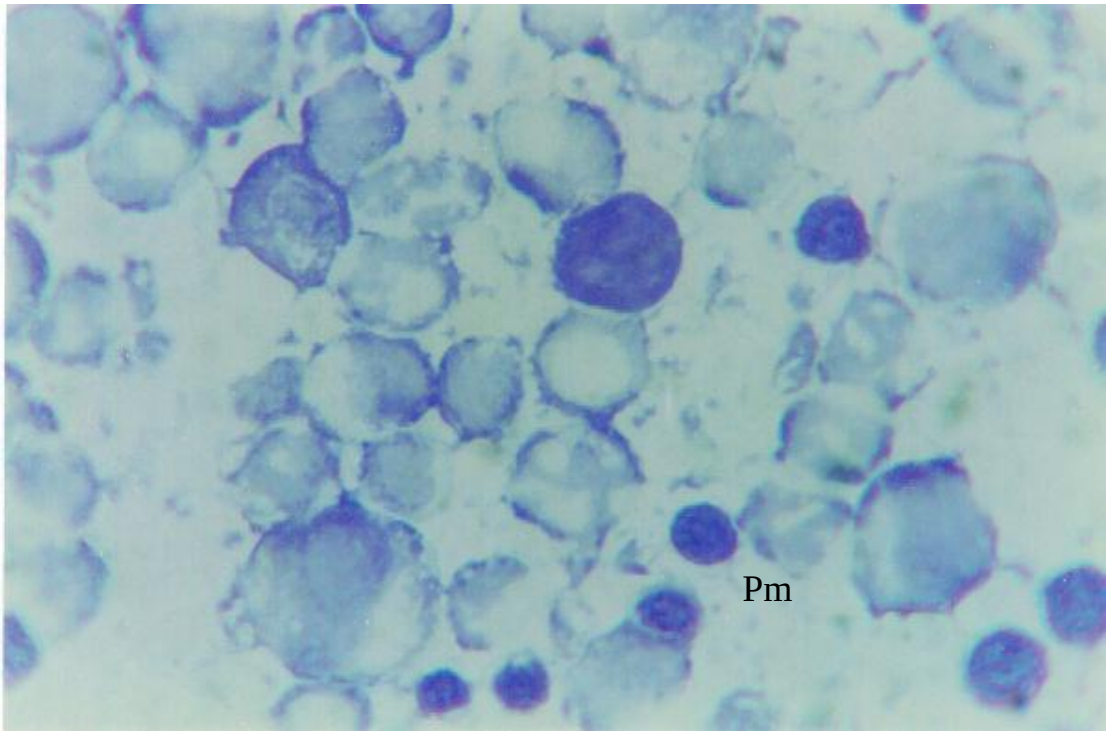
- Original magn. X 1000.



- MBB reaction.

- Fig (32): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined one day post-irradiation, showing a slight decrease in the reactivity of the cells inside the damaged marrow environment. Extracellular protein material (arrows) appeared in the field due to cellular damage.

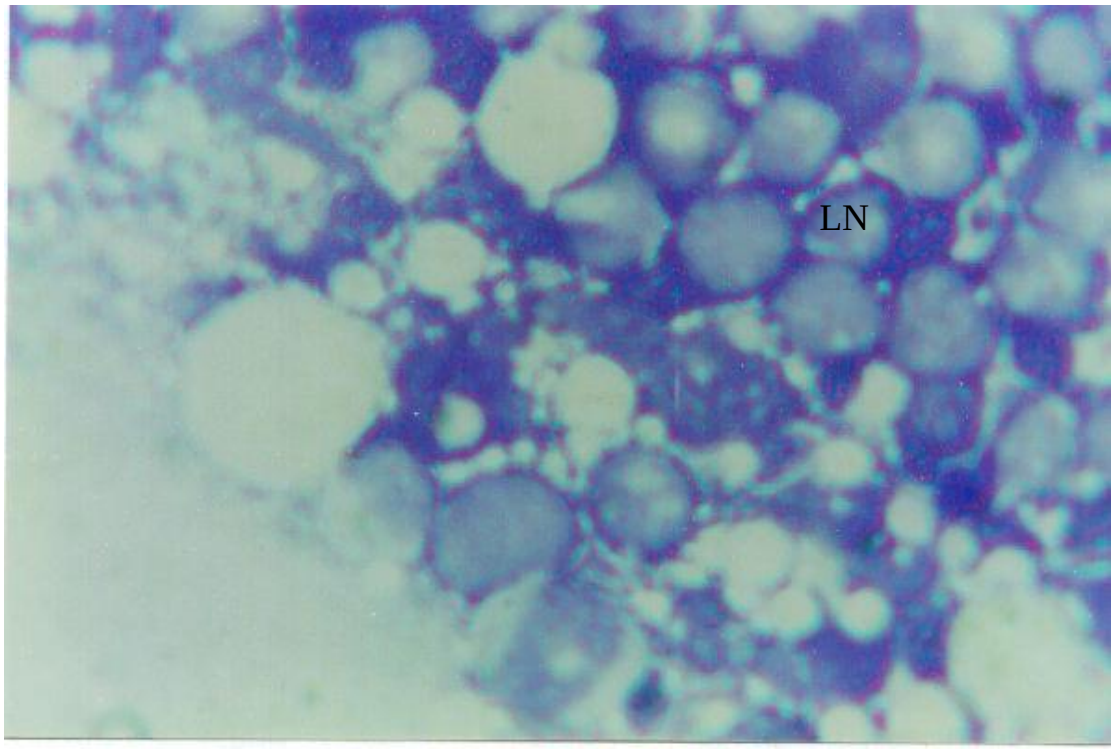
- Original magn. X 1000.



- MBB reaction.

- Fig (33): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined three days post-irradiation, showing a more decrease in the protein content inside the highly destructed nucleated cells. Except few cells of the polymorphs (Pm) showing intense MBB reaction.

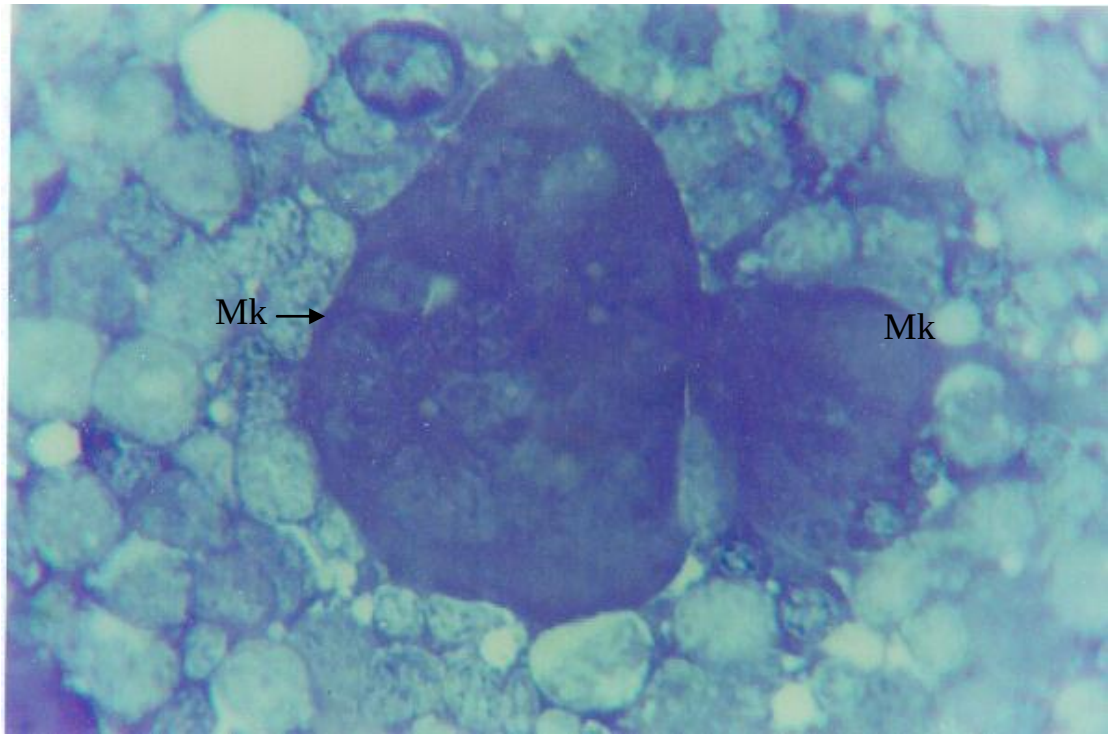
- Original magn. X 1000.



- MBB reaction.

- Fig (34): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined seven days post-irradiation, showing a high increase in the protein content inside the cells of leucocytic series and late normoblasts (LN).

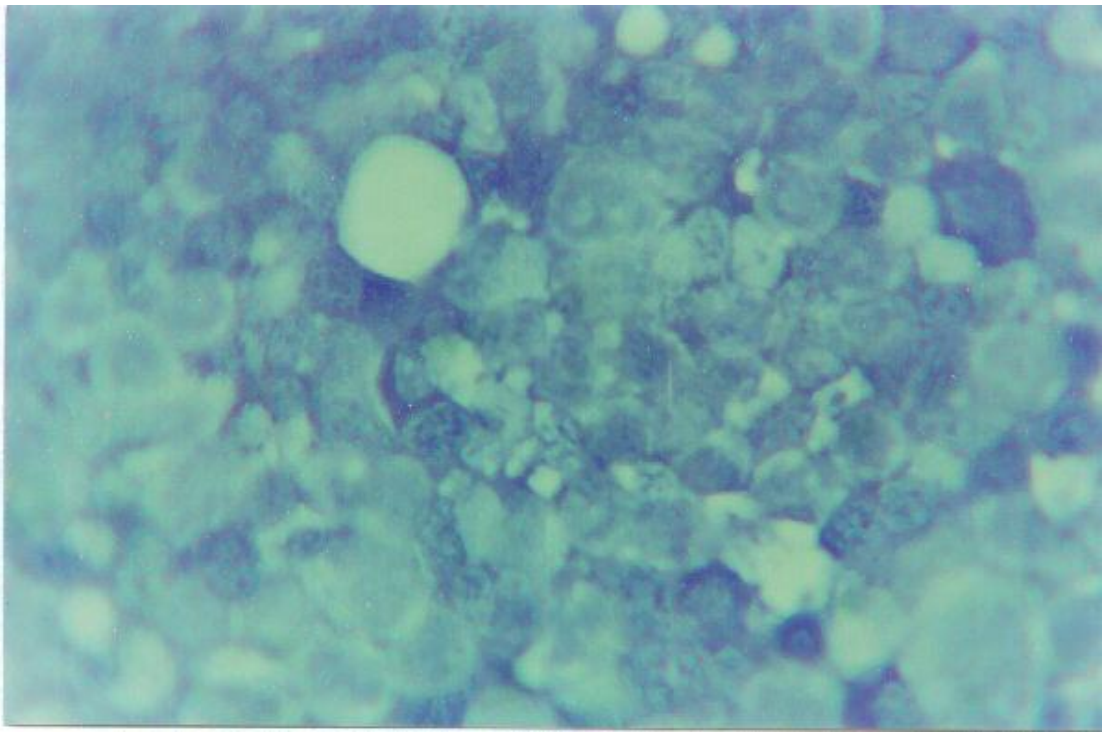
- Original magn. X 1000.



- MBB reaction.

- Fig (35): Bone marrow smear of rat irradiated at 6Gy gamma rays and examined fifteen days post-irradiation, showing the marrow cells with variable protein intensities nearly similar to that of the normal cells. In the field, appears two megakaryocytes (Mk) exihibiting high protein positive materials.

- Original magn. X 1000.



- MBB reaction.

- Fig (36): Bone marrow smear of rat irradiated 6Gy gamma rays and examined thirty days post-irradiation, showing protein staining approaching the normal picture in the cells of normal rats.

- Original magn. X 1000.