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Grambling, Louisiana 71245

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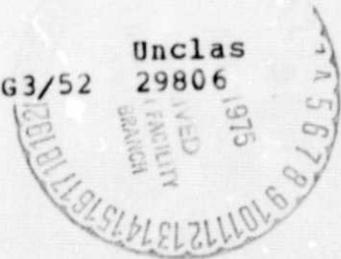
\*See comments on nature of funding and of this report in preface, p.i.

(NASA-CR-143262) LYMPHOID CELL KINETICS  
UNDER CONTINUOUS LOW DOSE-RATE GAMMA  
IRRADIATION: A COMPARISON STUDY Final  
Technical Report (Grambling State Univ.,  
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## PREFACE

This research project was jointly funded by NSF (Grant GB 29136) and NASA (Grant NGR-19-011-008) during the first two years of investigation. The research was initiated at The Argonne National Laboratory where the principal investigator was a participant in The Argonne Faculty Research Participation Program, summer, 1971. Research support during the third year of investigation was sponsored by NASA (Grant NGR-19-011-008, Supplement #1). During the final year of this study, research support was granted by NASA (Grant NSG 9014).

Findings during the first three years of investigation have been reported as a final report as they pertained to NASA Grant NGR-19-011-008, dated October 4, 1974. Findings reported here pertain to NASA Grant NSG 9014.

Lymphoid Cell Kinetics Under Continuous Low  
Dose-Rate Gamma Irradiation - A Comparison Study

by

Bessie Ruth Foster

Scope, Methods, and Findings

The research involved making a comparison study of the effects of continuous low dose-rate gamma irradiation on cell population kinetics of lymphoid tissue (white pulp) of the mouse spleen with findings as they relate to the mouse thymus.

Experimental techniques employed included autoradiography and specific labeling with tritiated thymidine (TdR-<sup>3</sup>H).

The problem studied involved the mechanism of cell proliferation of lymphoid tissue of the mouse spleen and thymus under the stress of continuous irradiation at a dose rate of 10 roentgens (R) per day for 105 days (15 weeks). The aim was to determine whether or not a steady state or near-steady state of cell population could be established for this period of time, and what compensatory mechanisms of cell population were involved.

Exposure of the test animals to continuous irradiation was carried out at The Argonne National Laboratory. Subsequent studies on these subjects are described below.

Unirradiated animals of the same age, strain, and sex were processed in a similar manner as irradiated mice. These mice served as controls.

Each data point on any given graph represents an average on 4 animals, and at least 1000 thymus or spleen cells (white pulp) were counted and categorized per microscopic examination per animal.

Phase one of the study involved irradiation of one hundred, 29-day old BCF<sub>1</sub> male mice at a dose rate of 10R per day in order to obtain some information pertaining to changes in tissue weight. Four mice were removed from the irradiation unit, and sacrificed one hour following an intraperitoneal injection of TdR-<sup>3</sup>H at various time intervals up to 105 days. Standard techniques were employed to dissect lymphoid tissue, obtain tissue weights, cell counts, autoradiographs, etc.

#### Changes in Tissue Weight

Changes in lymphoid tissue weight under continuous gamma irradiation at 10R per day were examined in 4-week old BCF<sub>1</sub> male mice at 0, 1, 2, 3, 4, 5, 6, 7, 10, 14, 18, 21, 24, 28, 35, 42, 49, 56, 63, 70, 77, 84, 91, 98, and 105 days. Figure 1 shows that initially there was an "outburst" of growth in the thymus indicated by a sharp increase in tissue weight. This finding was interpreted as a "compensatory response" to the initial radiation stimulus. Thereafter, weights of irradiated thymuses seemed to parallel those of controls, albeit at a reduced level, up to 49 days of irradiation at which time there was an increase in the weight of irradiated thymuses. A t-Test on these data indicated that the difference was significant at the 5% level. A similar finding occurred at day 63--however, in reverse order; that is, there was a significant decrease in thymus weights among irradiated mice. This occurrence was followed by results that approximated each other in irradiated and control thymuses up to day 84. At this time there was a significant decrease in thymus weights, among

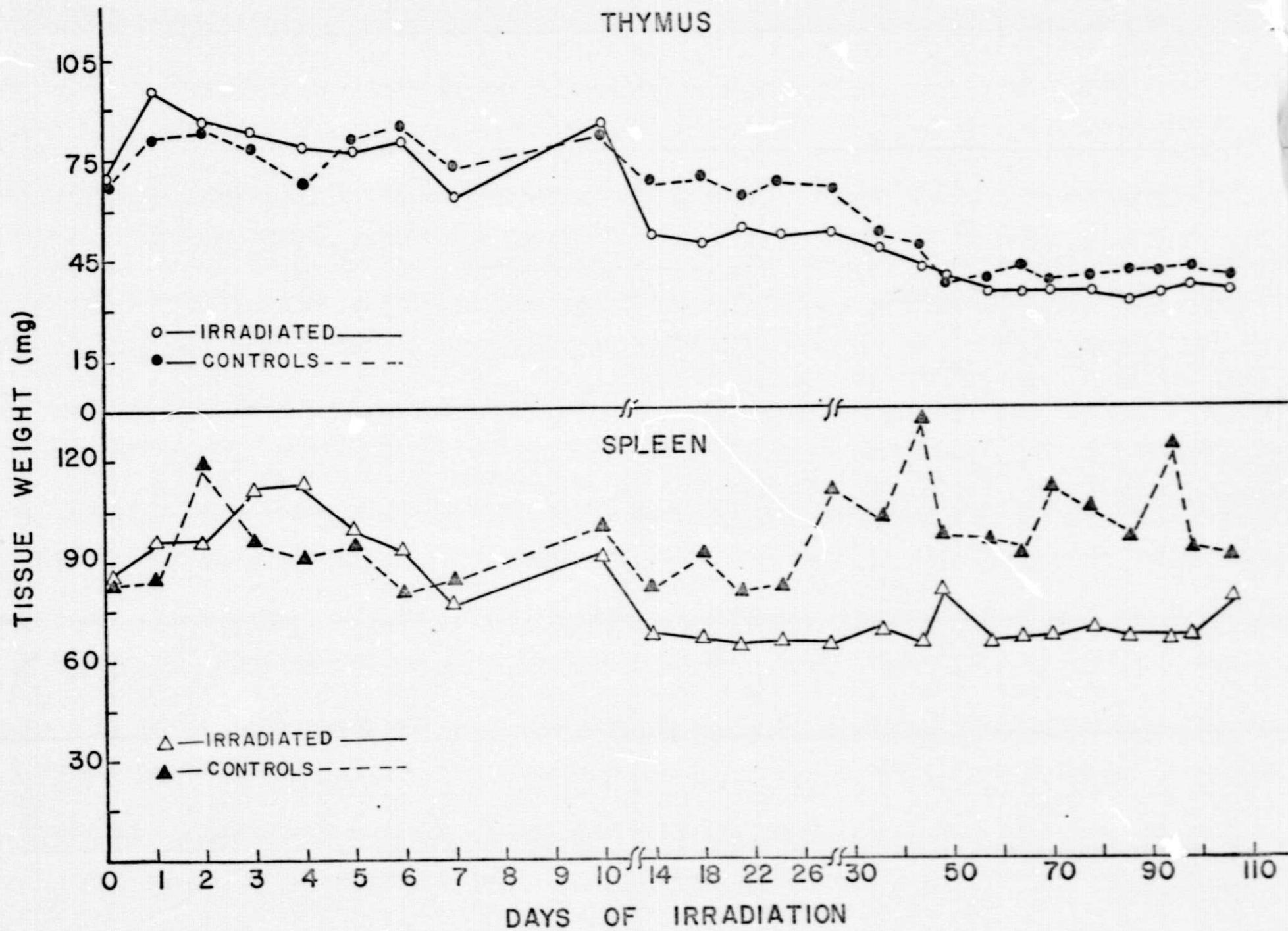


Figure 1

irradiated mice compared to controls up to day 48, with a steady state being achieved again by day 105, the termination day of radiation exposure for this study.

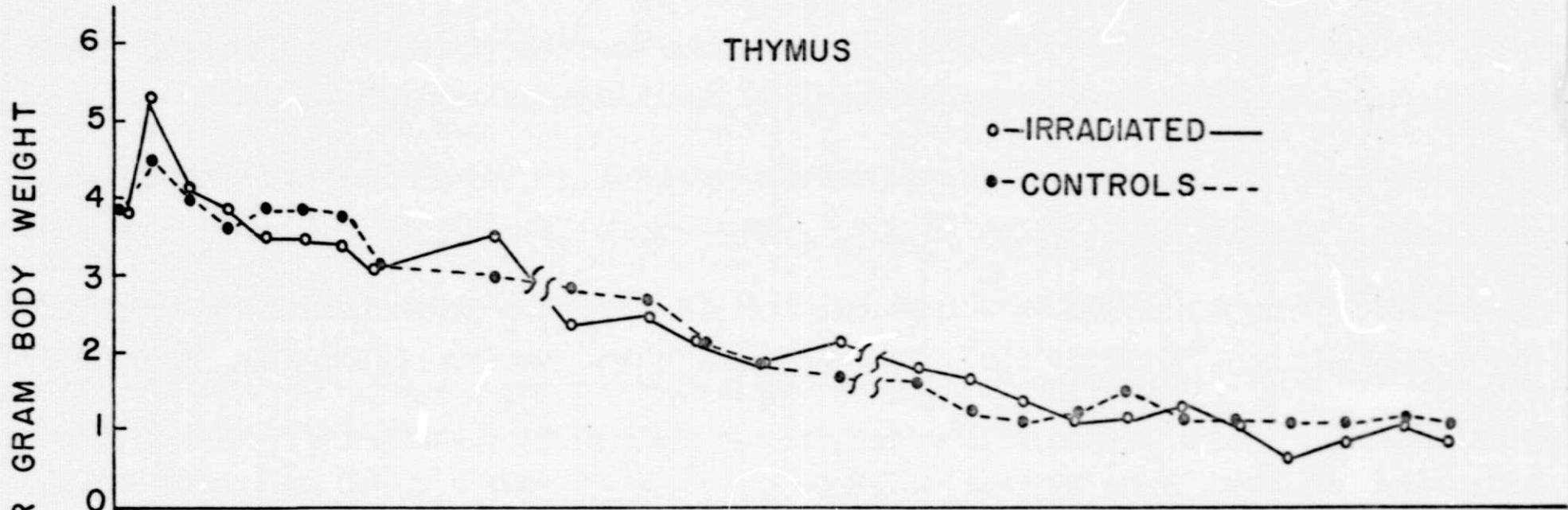
On the basis of thymus weights and tests of significance carried out on these data, it was concluded that the thymus in BCF<sub>1</sub> male mice seemed to maintain a steady or near-steady state of tissue growth up to 84 days of irradiation at a dose rate of 10R per day. At this time there was a "breakdown" in thymus tissue growth among irradiated mice for about two weeks with a steady state being achieved again by day 105 albeit at a much lower level than what appears to be normal.

As in the thymus, the spleen also exhibited an "outburst" of growth after the initial radiation treatment. Following this, there was a significant difference among weights in irradiated spleens versus controls for about 18 days of irradiation with higher weight values shifting from controls to irradiated and back to controls. By day 21, the spleen had achieved a steady-state of growth, for there was no significant difference among tissue weights in the two groups. However, the steady state phenomenon was very short-lived lasting for only a few days. At day 28 through 42, there was a significant decrease in weights among irradiated spleens, followed by another short-lived period of steady-state growth for about a week encompassing day 49. From day 56 through 98, there was another decrease in tissue weights among irradiated spleens. A t-Test on these differences were significant at the 5% level. By day 105, irradiated spleens had again achieved a steady state of tissue growth.

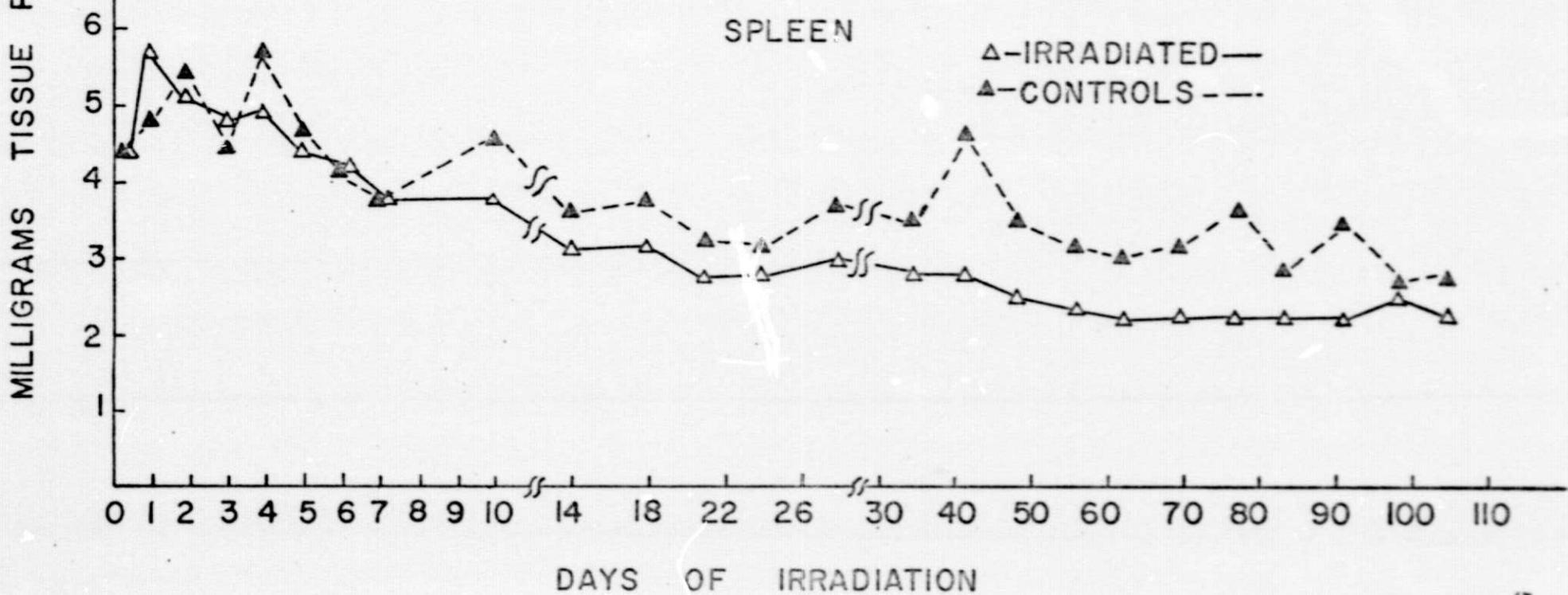
Weight changes in thymus and spleen tissue per gram body weight were examined under continuous irradiation at various time intervals from day zero to day 105. Figure 2 illustrates changes that occurred. As reported

# RADIATION EFFECT ON LYMPHOID TISSUE WEIGHT

## THYMUS



## SPLEEN



above, there was an "outburst" of cellular proliferation in irradiated tissue at the onset of irradiation in both the thymus and spleen. This finding is interpreted as a "compensatory response" to the initial radiation treatment. Thereafter, the weights in control and irradiated thymuses/gram body weight paralleled each other throughout the remainder of the 105 day irradiation period. However, this was not the case with the spleen. From about day 35 through day 91, there was a marked decrease in the weight of the spleen per gram body weight in irradiated animals. These findings are consistent with findings of other investigators. Neary, Munson and Mole (1957) observed that under continuous gamma irradiation, the weights of the mouse thymus and spleen were maintained at a dose rate of 16R per day for 15 weeks, the spleen being affected more. At 40 rads per day, Lamerton, Pontifex, Blackett and Adams (1960) and Lamerton (1966) observed that peripheral blood lymphocyte counts in rats fell by day 20, then rose, and recovery with normal spleen weight was maintained with animals surviving up to 30 weeks. In splenectomized rats, the rise after 20 days was not seen, and blood lymphocyte levels stabilized at a lower level than normal. Brecker, Brambel and Brambel (1964), observed low peripheral blood lymphocyte counts, collapse of splenic lymphoid follicles, and marked splenic erythropoiesis in rats after 70 rads per day for 12 weeks.

It is of interest to note that even though the radiation dose rate which we employed was very low (10R/day), we observed similar weight changes in the thymus and spleen that other investigators observed at higher dose rates. Thus, lymphatic tissue is among the most radiosensitive tissue of the body as reported by Trowell (1952, 1961) and Vos (1967).

Findings discussed to this point suggest that on the basis of tissue weight, lymphoid tissue in BCF<sub>1</sub> male mice seemingly maintains a steady or near-steady state of cellular proliferation under continuous irradiation for up to 105 days, with intermittent periods of "collapse". These phenomena imply a bi-phasic response of tissue growth to the radiation dose administered.

The next phase of investigation was to examine the effect of continuous irradiation on the distribution of various lymphoid cell types in the thymus and spleen to ascertain some idea of the compensatory mechanisms involved in the steady or near-steady state phenomenon that is established.

#### Distribution of Lymphoid Cell Types

Cell types considered here were of three classes: PAS-positive reticular cells, non-PAS-positive reticular cells and lymphocytes.

Figure 3 illustrates the distribution of PAS-positive reticular cells in the thymus and spleen under continuous irradiation. Although the total number of PAS-positive cells observed was small, a significant increase in their abundance occurred in irradiated thymuses during the latter phase of the irradiation period. There was also an increase in the proportion of PAS-positive cells in the irradiated spleen, but only during the initial phase of the irradiation period.

PAS-positive reticulum cells in the thymic cortex and mitoses in the lymphoid cells in contact with the PAS cells have been observed (Goodwin, 1939; Gordon, 1955; Metcalf and Ishidate, 1962; Metcalf, 1964). These cells are reported to be most marked in the thymuses of non-neoplastic, high leukemia AKR mouse strains, and to a lesser degree in the thymuses of other mouse strains. In the thymus of high-leukemia AKR mouse strains,

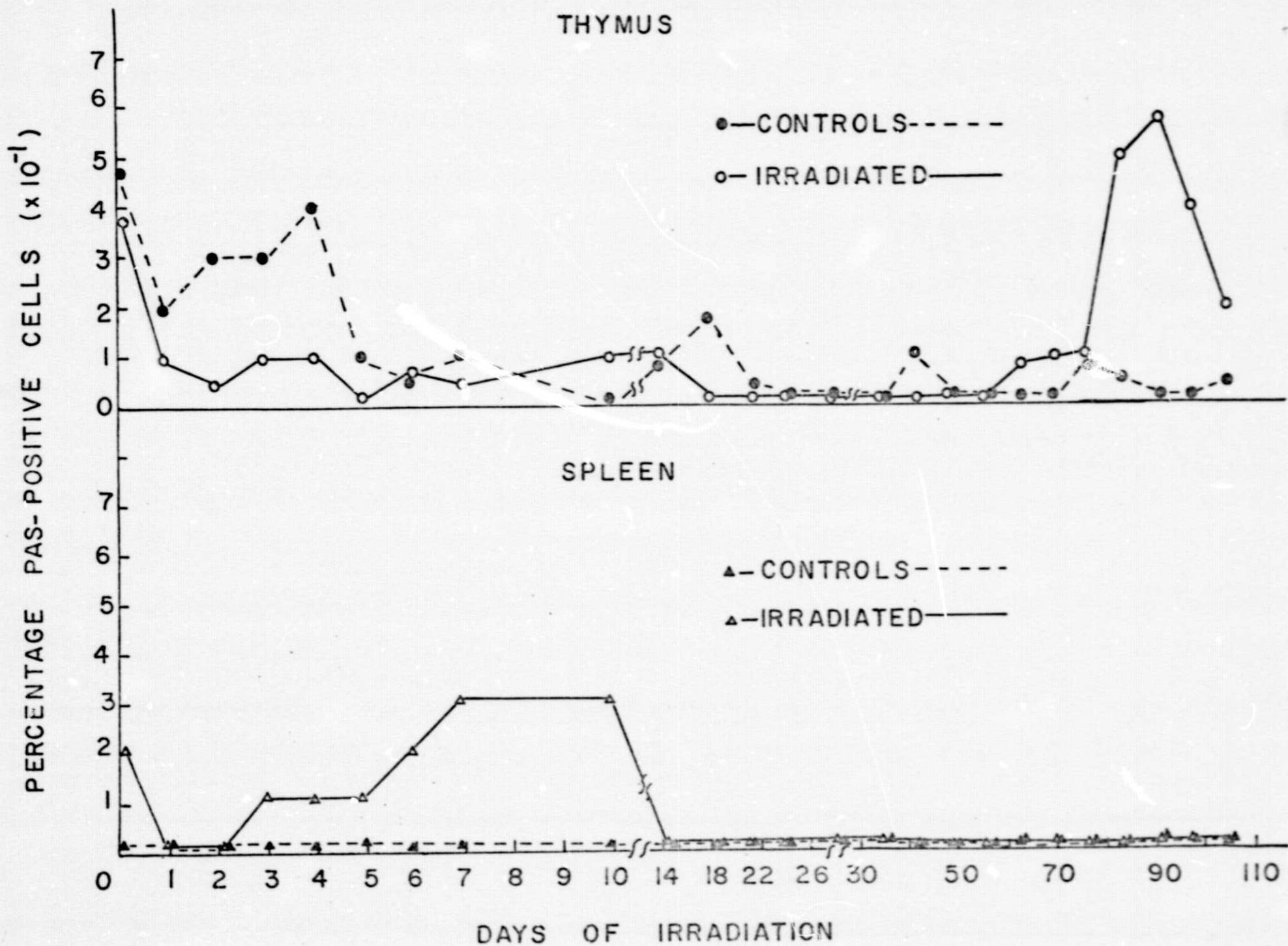


Figure 3

there were three times as many mitoses in lymphocytes than elsewhere in the thymus (Metcalf and Ishidate, 1962; Metcalf, 1964). These changes were interpreted as a "triggering process" induced by PAS-positive reticulum cells stimulating mitosis among lymphocytes coming in contact with them, and thereby regulating lymphopoiesis.

The absence of PAS-positive cells in the spleen from about two weeks of irradiation throughout the remainder of the 105-day irradiation period suggests that an increased stimulus for mitotic activity as evidenced by the presence of PAS-positive cells was not operative in splenic lymphoid tissue at a similar time interval nor to the same extent as that exhibited in the thymus.

Data on non-PAS-positive reticular cells are presented in Figure 4. These data illustrate that the proportion of non-PAS-positive reticular cells was generally smaller in irradiated thymuses and spleens at the onset of irradiation, compared to non-irradiated controls. After the first few weeks of irradiation, there was an increase in the proportion of non-PAS reticular cells in the irradiated thymus. However, this occurrence was not observed in the spleen until about 6 weeks of irradiation, and the increase in the proportion of non-PAS reticular cells in irradiated spleens was only slightly greater than that observed in comparable controls.

Since reticular cells of lymphoid tissue are precursor cells which give rise to lymphocytes, these findings suggest that another compensatory mechanism which serves to re-establish the near-steady state of cellular proliferation in the thymus and spleen under continuous irradiation is a state of "maturation arrest" among non-PAS-positive reticular cells. That is, a greater proportion of the lymphoid cell population remains in the "progenitor" or "precursor" state under continuous irradiation, thereby,

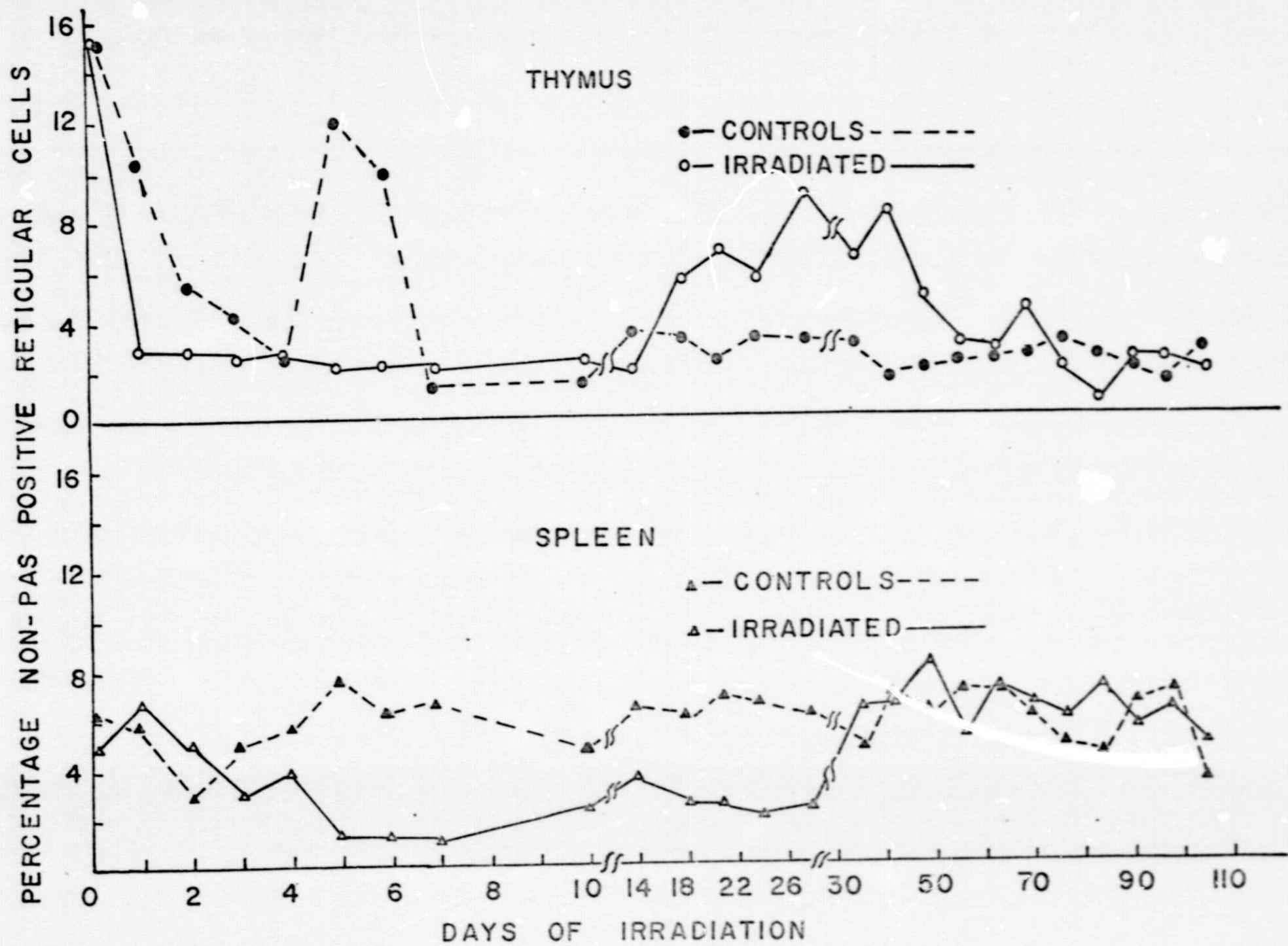


Figure 4

producing cells which compensate for those that are damaged or destroyed by radiation.

As with the findings relative to PAS cells in the thymus and spleen, the increase in the proportion of non-PAS reticular cells was observed at different time intervals and to a lesser extent in irradiated spleen tissue compared to the thymus.

The proportion of lymphocytes in the thymus and spleen under continuous irradiation is illustrated in Figure 5. This figure shows that the percentage of lymphocytes in both thymus and spleen tissue averaged about 95% in irradiated and in control animals. Therefore, there was no significant change in the proportion of lymphocytes under continuous irradiation.

#### The Cell Cycle

On the basis of lymphoid tissue weights, it was demonstrated that a seemingly steady state of cellular proliferation was achieved by about three or four weeks of continuous irradiation. In order to determine whether or not there were any changes in the generation time of lymphoid cells, a group of 40, four-week old BCF<sub>1</sub> male mice was exposed to continuous irradiation at a dose rate of 10R per day for 4 weeks, removed from the irradiation unit, injected intraperitoneally with TdR-<sup>3</sup>H, and sacrificed at various time intervals ranging from 0.5 to 30.0 hours following TdR-<sup>3</sup>H. Lymphoid tissue was dissected, fixed, processed through autoradiography, developed, stained, examined microscopically, and labeled mitoses scored at each sacrifice interval using routine histological and autoradiographic procedures.

Forty unirradiated mice of the same age, sex, and strain were processed similarly, these mice served as controls.

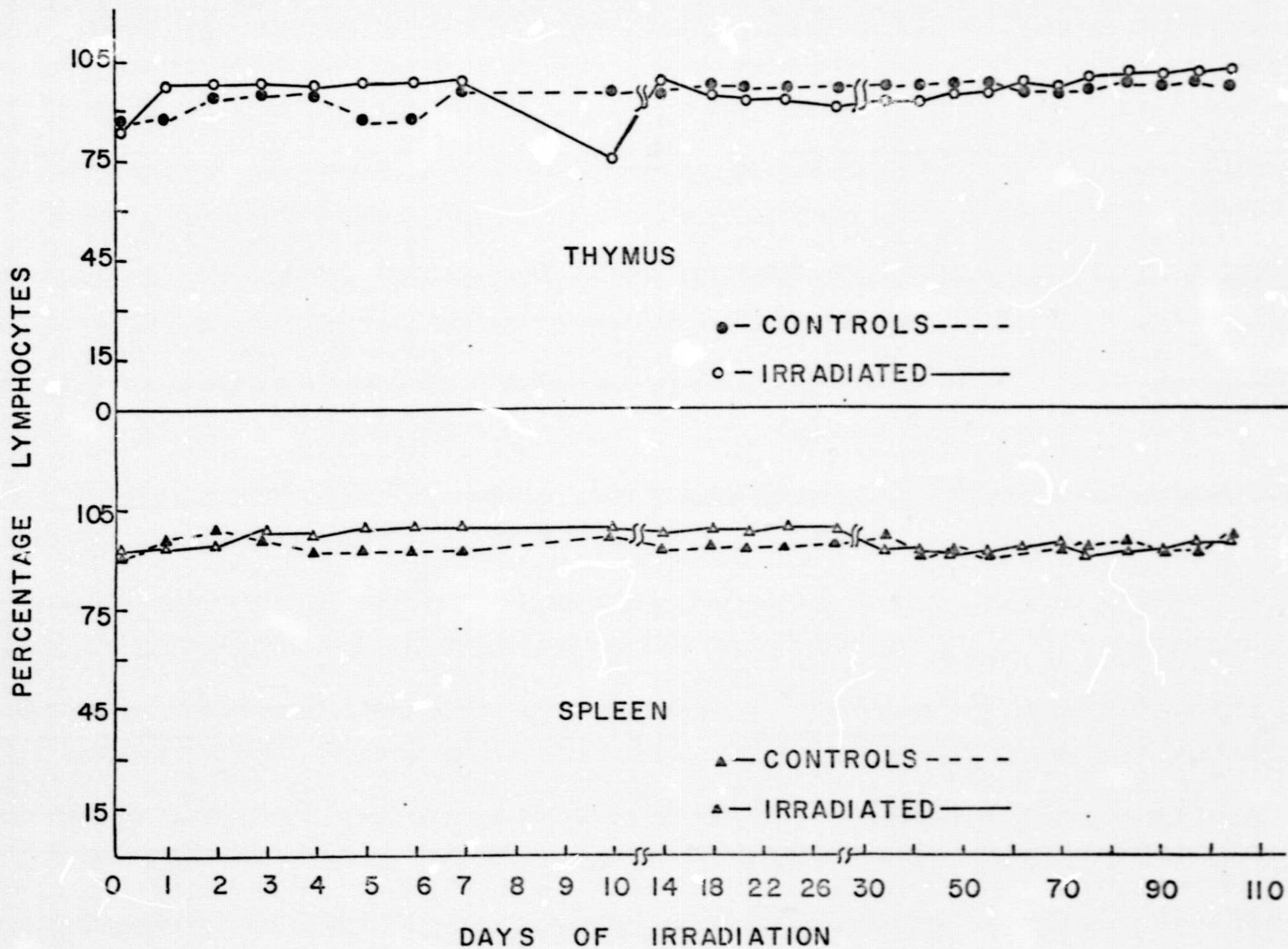


Figure 5

Two hundred mitotic figures were scored at each sacrifice interval, and data relative to the thymus were graphed to form labeled mitosis curves, as shown in Figure 6.

Although there was no well-defined descending limb of the first wave of mitosis nor was there a well-defined second wave in irradiated nor in control thymuses, it was possible to roughly approximate a cell cycle time of about 10.5 hours in both groups. It was concluded, therefore, that changes in the cell cycle time in thymuses irradiated at 10R per day were not contributing compensatory mechanisms.

Theoretically, all cells in DNA synthesis at the time of TdR-<sup>3</sup>H injection should have been labeled and should have passed through mitosis to give 100% labeling (Johnson, 1961). However, in the present study labeling reached only about 98% on the ascending limb of the initial wave in irradiated thymuses and about 96% in control thymuses at 5 hours and 3 hours after TdR-<sup>3</sup>H, respectively. Less than 100% labeling during the first wave of the cell cycle has been reported in spontaneous breast cancer (Mendelsohn, Dohan and Moore, 1960), transplanted tumors (Steel, Adams and Barrett, 1966), and the regenerating liver (Fabrikant, 1964; 1967a); this may be due, in part, to the spread in the duration of the G<sub>2</sub> + M/2 complex (Fabrikant, 1967a) and the failure of a proportion of labeled cells (false negatives) lying deep in a section to give a radiographic image because of the very short range of tritium beta particles (Johnson, 1961; Fabrikant, 1967a; Fry, personal communication).

The mean duration of the S phase in both irradiated and control thymuses was approximated to be about 6.5 hours. Since the seemingly descending limb of the initial wave of both curves is less steep than the rise, the S phase in individual cells (reticular cells, large, medium, and small

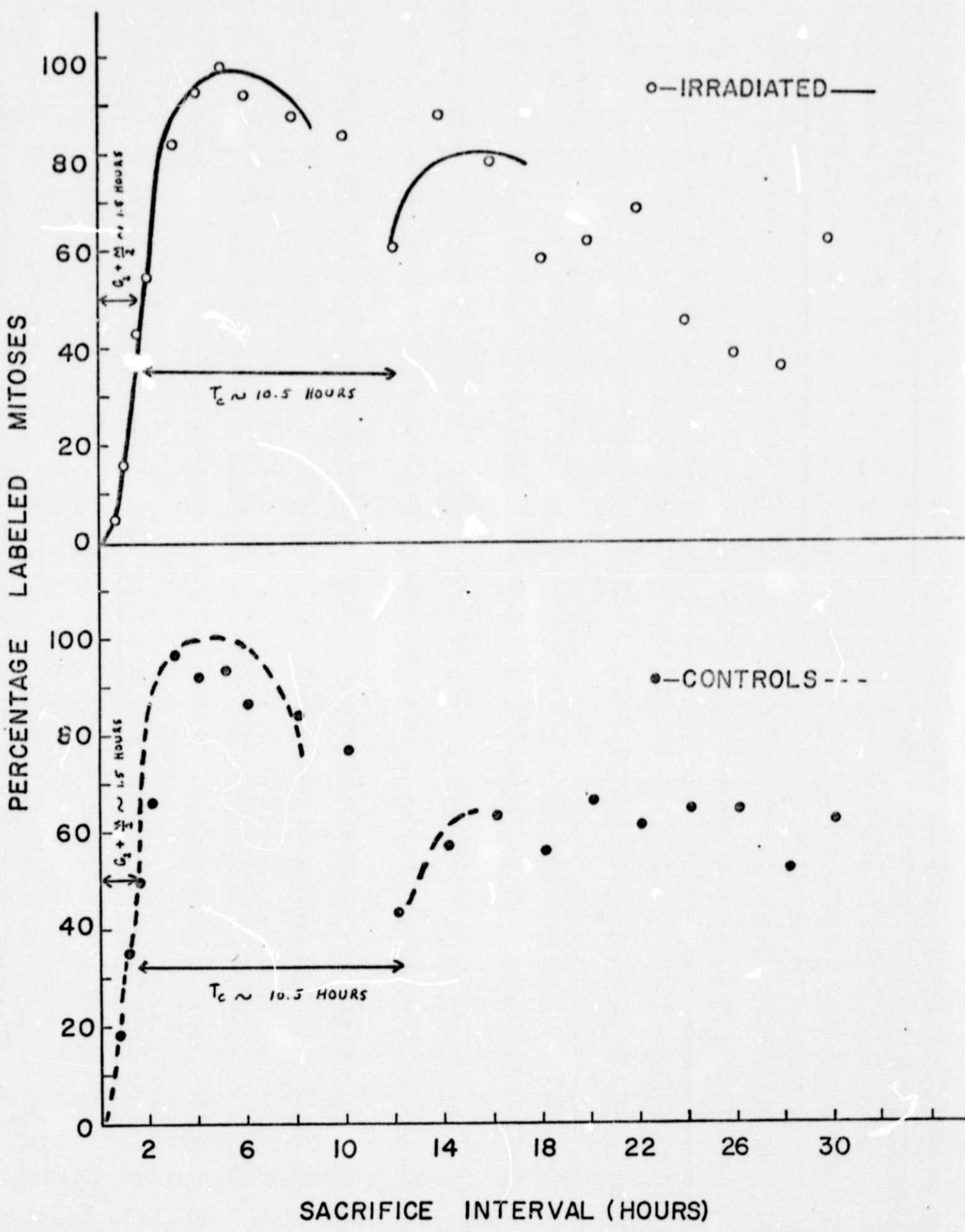


Figure 6

lymphocytes) is of unequal lengths. The low labeling among mitotic figures following the peak is possibly due to cells which were in the G<sub>1</sub> phase at the time of labeling. The variability in the duration of the G<sub>1</sub> phase must be extremely great, which partly accounts for the absence of a well-defined peak for the S phase of the daughter cells.

Curves of this type are characteristic of several tissues such as the small intestine (Quastler and Sherman, 1959), bone marrow (Cronkite, Bond, Fliedner, and Rubini, 1959), tumors (Mendelsohn, et al., 1960; Mendelsohn, 1962; 1963), hair follicles (Cattaneo, Quastler, and Sherman, 1961; McCarter and Quastler, 1962; Greim, 1966), skin (Gelfant, 1963), and regenerating liver (Fabrikant, 1967a; 1967b; 1967c; 1968a; 1968b).

Furthermore, the radiation response of a cell system may very well be more marked in autoradiographic studies due to intranuclear radiation effects from incorporated TdR-<sup>3</sup>H in cells preparing for division (Painter, Drew and Hughes, 1958; Drew and Painter, 1959; Johnson and Cronkite, 1959; Wimber, 1959; 1964; Post and Hoffman, 1961).

Another factor which should be taken into account is that the thymus is a multicompartmental cell system with at least 4 major cell types, each with a different generation time. Therefore, the findings reported here are, at best, a rough approximation of the cell cycle time.

As far as a cell cycle time for spleen tissue is concerned, a labeled mitosis curve was not practical in that mitotic figures are difficult to observe. However, it was possible to calculate the maximum cell cycle time for both thymus and spleen cells using the following relationship:

$$T_c = T_s / LI$$

where T<sub>c</sub> is the duration of the cell cycle, T<sub>s</sub> is the duration of DNA syn-

thesis and LI is the labeling index. Maximum cell cycle times among thymus and spleen lymphoid cells are shown in tabular form in Table 1. It should be emphasized here that the calculated values for maximum cell cycle times found in Table 1 are based on the entire population of cells rather than individual cell types. And the wide range of values may be due to the fact that there are two populations of lymphocytes, one relatively short-lived and the other long-lived (Ottesen, 1954; Hamilton, 1956; 1958). Nonetheless, it was possible to demonstrate that there was no appreciable decrease in the maximum cell cycle times among irradiated thymuses and spleens compared to comparable controls.

Since there was no apparent decrease in the generation time of thymus and spleen cells in irradiated mice compared to controls, the next line of investigation was to determine the distribution of lymphocyte classes, thymidine labeling, proliferative fraction, and relative proliferative capacity among each lymphocyte class in order to determine which cell type(s) was contributing most to the proliferative activity of the cell systems.

#### Distribution of Lymphocyte Classes and TdR-<sup>3</sup>H Labeling

Cells were classified on the basis of morphology and size as reticular cells, large, medium, and small lymphocytes.

Data on the distribution of reticular cells and various lymphocyte classes are illustrated relative to the thymus and spleen in Figure 7 and Figure 8, respectively. These figures show that there was generally an increase in the proportion of reticular cells and medium lymphocytes among irradiated thymuses compared to controls. Whereas, in the spleen, there was generally an increase in the proportion of reticular cells, and large and medium lymphocytes among irradiated animals.

TABLE 1

MAXIMUM CELL CYCLE TIMES (CALCULATED IN HOURS)  
AMONG THYMUS AND SPLEEN CELLS UNDER  
CONTINUOUS IRRADIATION

| Days<br>of<br>Irradiation | Thymus   |            | Spleen   |            |
|---------------------------|----------|------------|----------|------------|
|                           | Controls | Irradiated | Controls | Irradiated |
| 0                         | 163      | 73         | 125      | 361        |
| 1                         | 47       | 110        | 103      | 53         |
| 2                         | 67       | 53         | 77       | 232        |
| 3                         | 105      | 54         | 112      | 150        |
| 4                         | 144      | 61         | 155      | 191        |
| 5                         | 51       | 92         | 133      | 105        |
| 6                         | 44       | 99         | 112      | 59         |
| 7                         | 44       | 66         | 175      | 47         |
| 10                        | 49       | 138        | 111      | 59         |
| 14                        | 73       | 84         | 191      | 217        |
| 18                        | 141      | 138        | 114      | 232        |
| 21                        | 163      | 74         | 171      | 271        |
| 24                        | 125      | 114        | 148      | 309        |
| 28                        | 108      | 94         | 148      | 216        |
| 35                        | 93       | 135        | 141      | 197        |
| 42                        | 123      | 148        | 107      | 171        |
| 49                        | 123      | 88         | 141      | 155        |
| 56                        | 31       | 75         | 133      | 232        |
| 63                        | 33       | 94         | 191      | 186        |
| 70                        | 43       | 102        | 117      | 295        |
| 77                        | 44       | 87         | 108      | 283        |
| 84                        | 40       | 77         | 111      | 295        |
| 91                        | 57       | 33         | 127      | 241        |
| 98                        | 79       | 59         | 181      | 241        |
| 105                       | 171      | 71         | 181      | 241        |

# THYMUS

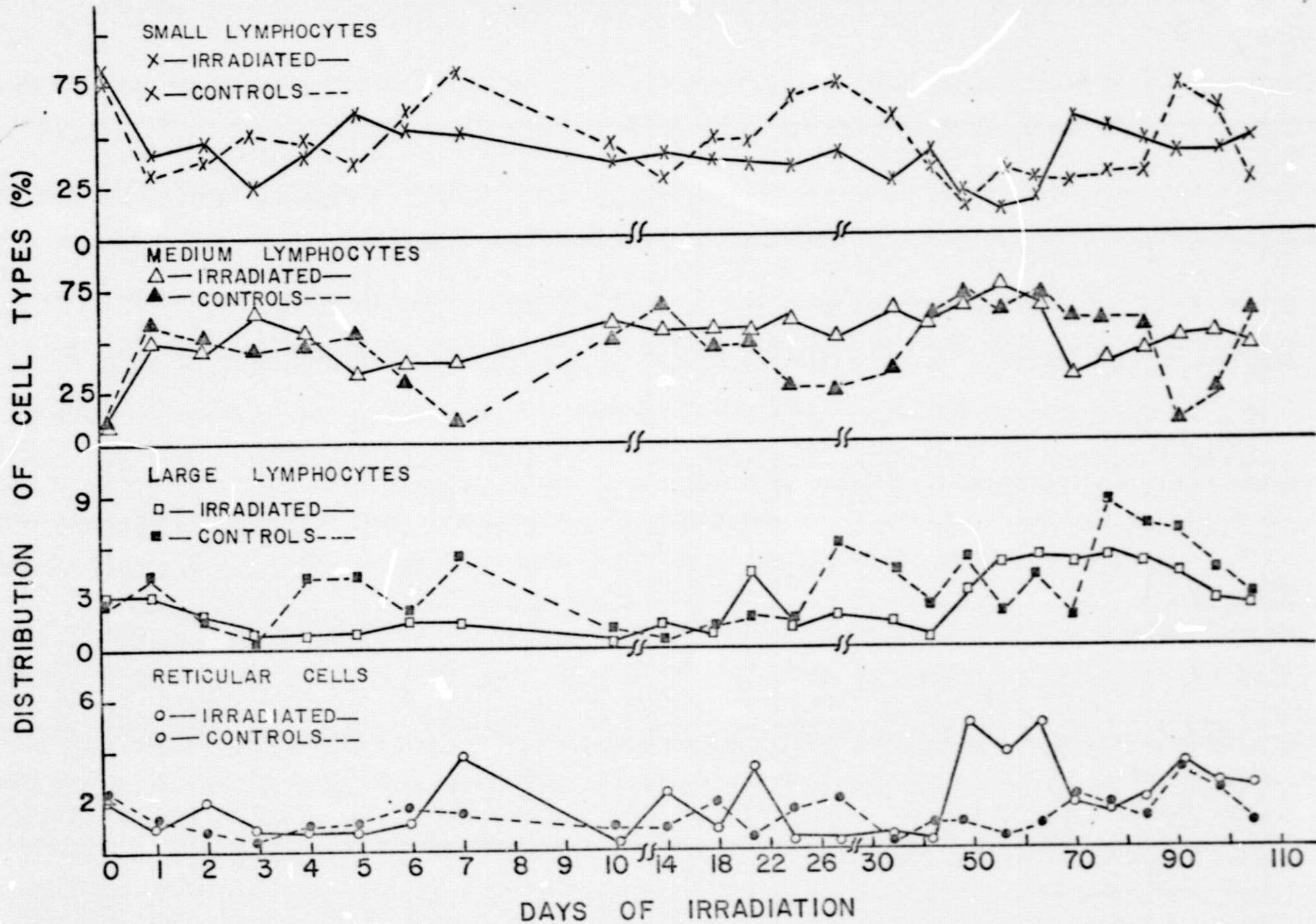


Figure 7

# SPLEEN

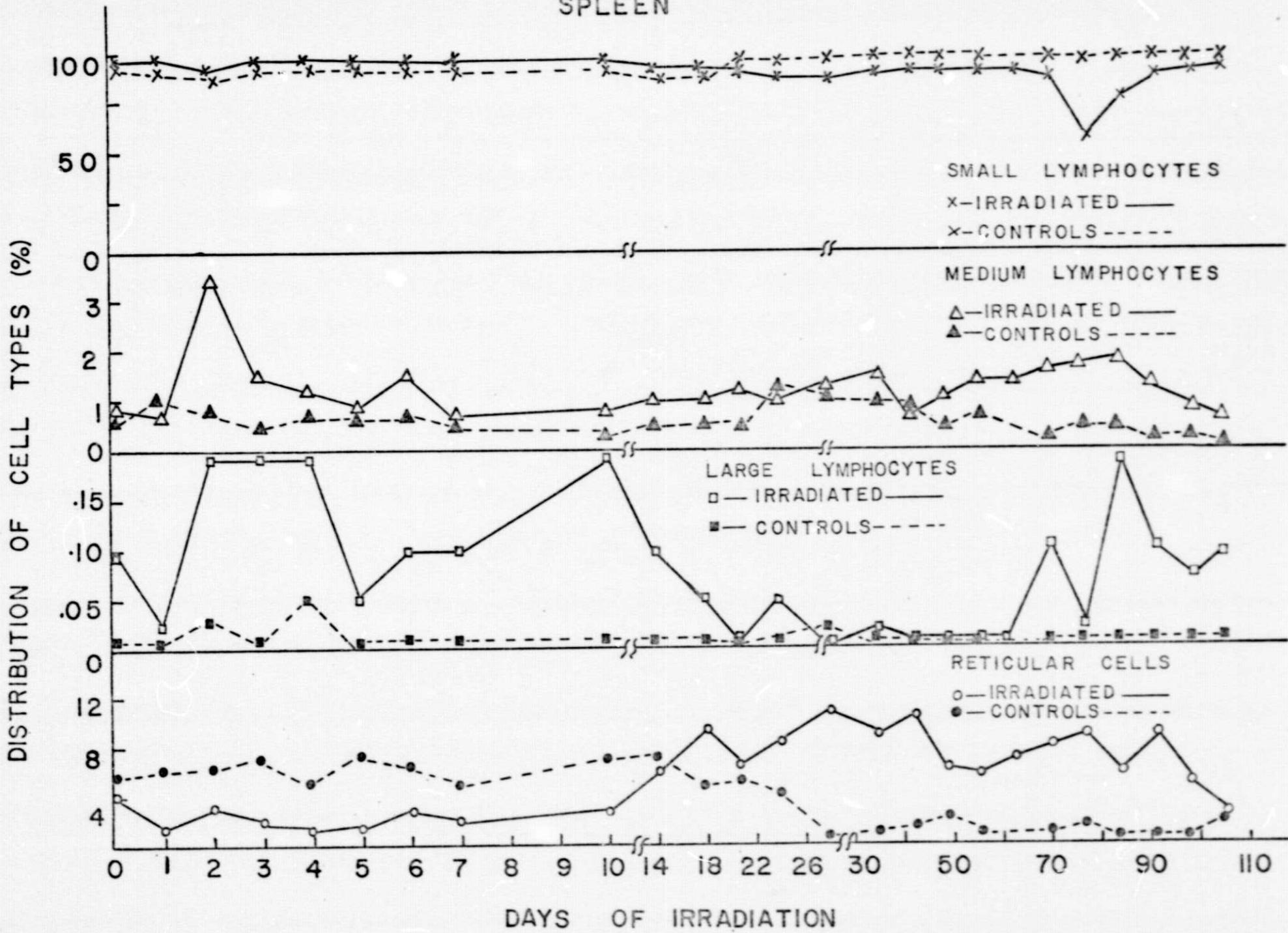


Figure 8

Since lymphopoiesis in lymphoid tissue appears to occur as a result of asymmetric division (Osgood, 1957; 1961) of reticular cells into lymphocytes of progressively decreasing sizes (Leblond and Sainte-Marie, 1960; Yoffey, 1960; Yoffey, et al., 1961), an additional compensatory mechanism in both the thymus and the spleen, under continuous irradiation, seems to be an increase in the "proportion of precursor cells."

Because of the relative numbers of medium lymphocytes along with a general increase in the proportion of this lymphocyte class in both the thymus and spleen suggested that the medium lymphocyte was contributing an appreciable amount to the proliferative activity of the cell system.

Thymidine labeling among various lymphocyte classes and in the total population is illustrated in Figures 9-11.

In the thymus, there was an increase in labeling among the medium lymphocyte class at the beginning of the radiation treatment in irradiated animals compared to controls. Also, there was a slight increase in labeling among all cell classes in irradiated thymuses at the end of the irradiation period.

In the spleen, there was an increase in labeling among irradiated reticular cells at the onset of irradiation, and a similar increase in labeling among irradiated reticular cells and medium lymphocytes near the end of the radiation exposure.

When data on labeling in the total population were examined, it was found that there was an increase in the average percentage of TdR-<sup>3</sup>H labeling among irradiated tissue in both the thymus and spleen near the end of the radiation treatment.

Since TdR-<sup>3</sup>H labeling suggests DNA synthesis, and since DNA synthesis suggests that cells are in preparation for division, these findings further

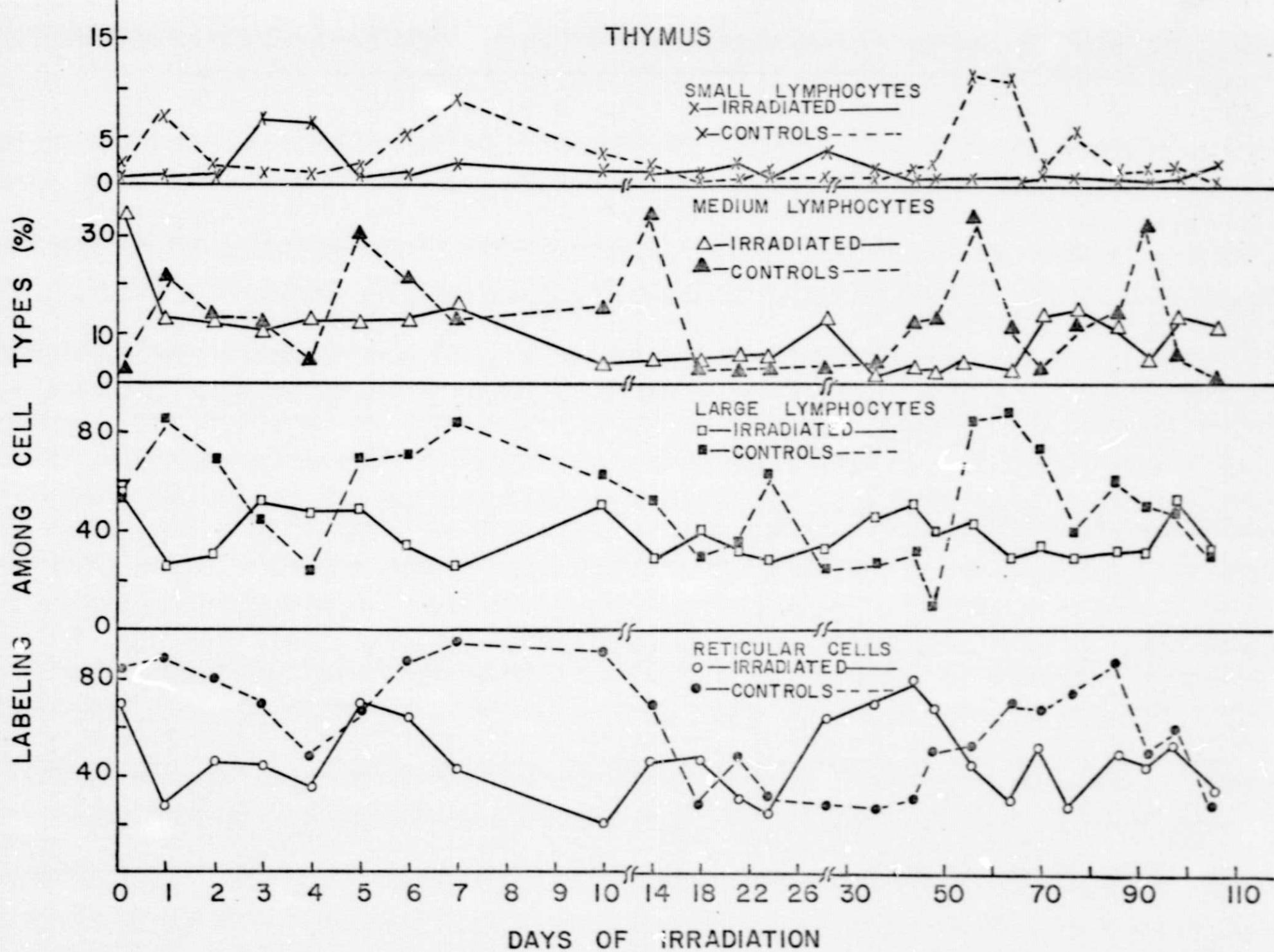


Figure 9

# SPLEEN

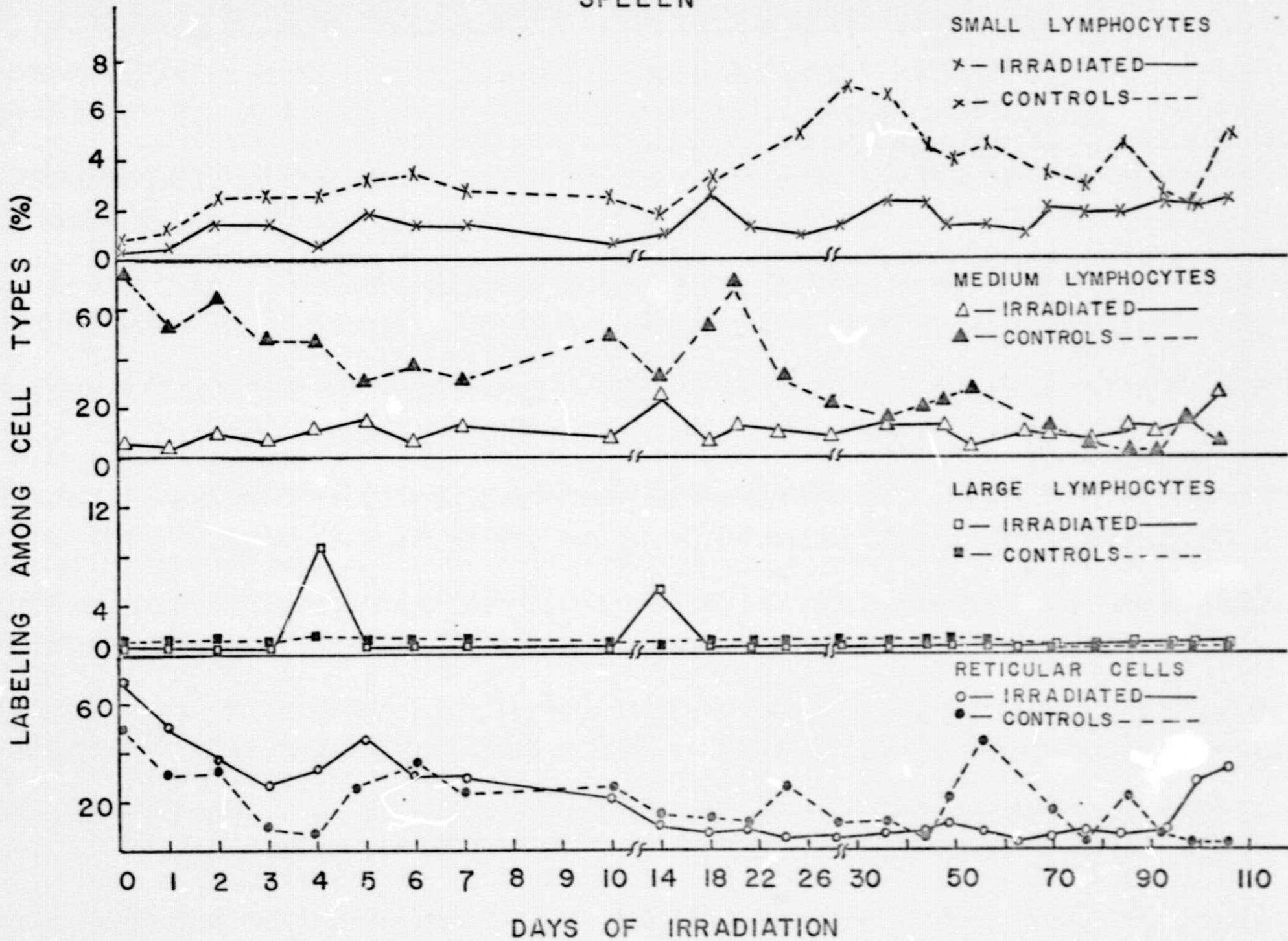


Figure 10

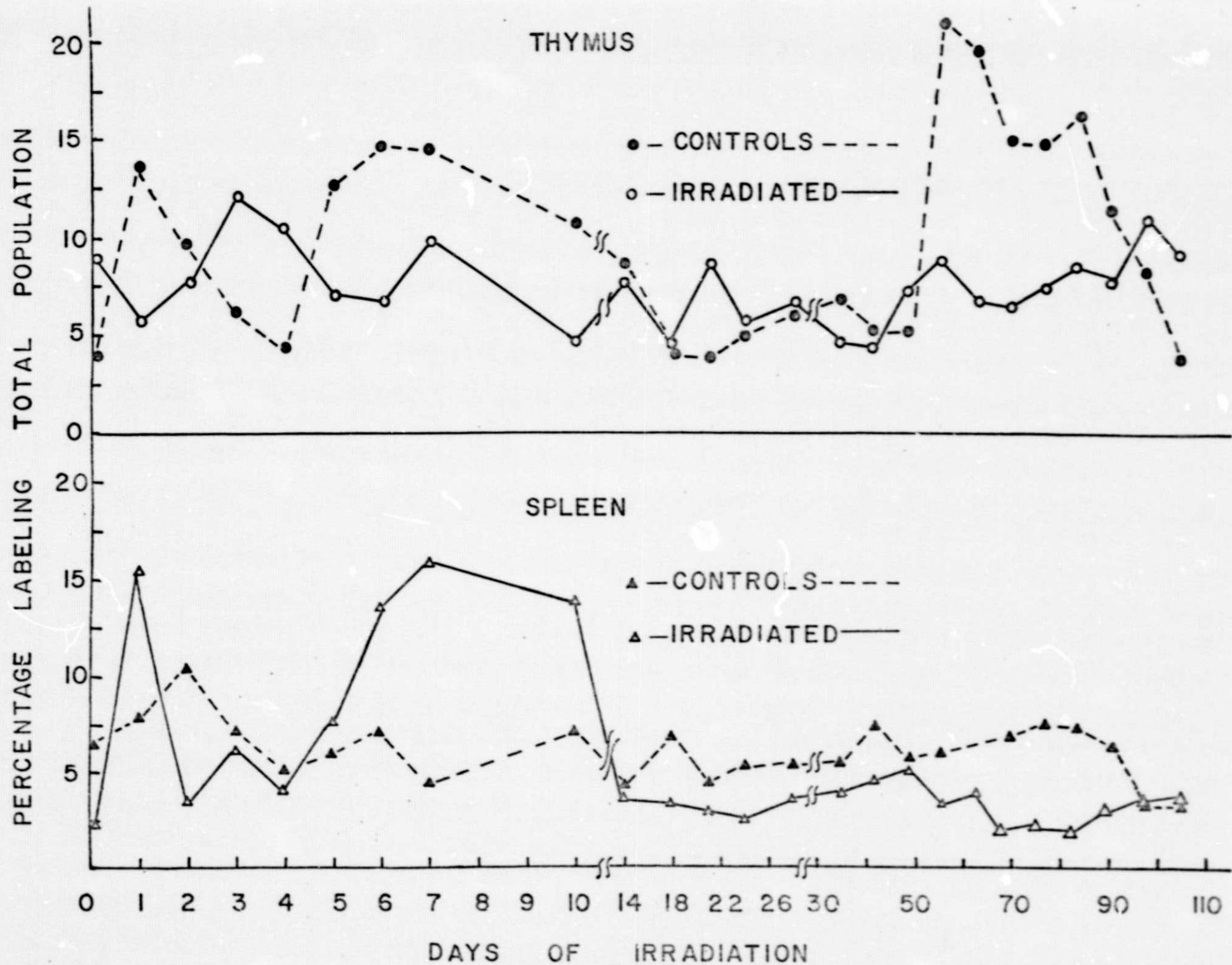


Figure II

support that the medium lymphocyte, in particular, is contributing an appreciable amount to the proliferative activity of the lymphoid cell system. Moreover, an increased tendency to proliferate, among certain cell types, is another compensatory mechanism that seems to be operative in maintaining a near-steady state in lymphoid tissue under continuous irradiation.

The final phase of this study was to determine the proliferative fraction and the relative proliferative capacity (RPC) among reticular cells and among the various classes of lymphocytes. That is to say, what proportion of a given cell population is in preparation for division, meaning the cell will perhaps divide, and what is the relative proliferative capacity which means how effective a cell class is in its capacity to proliferate compared to other cell classes.

#### Proliferative Fraction and Relative Proliferative Capacity

The proliferative fraction was calculated using the following relationship outlined by Lala and Patt (1966),

$$f = (N_S/N)/(N_S/N_G)$$

where  $N_S$  is the number of cells in DNA synthesis,  $N$  is the total number of cells in the population,  $N_G$  is the number of cells in cycle,  $N_S/N$  is the pulse labeling index, and  $N_S/N_G$  is the fraction of the proliferative population in DNA synthesis.

Data pertaining to the proliferative fraction among cell classes and in the total population are illustrated in Figures 12-14.

In the thymus, there was a general increase in the proportion of cells proliferating among irradiated medium and small lymphocytes at the onset

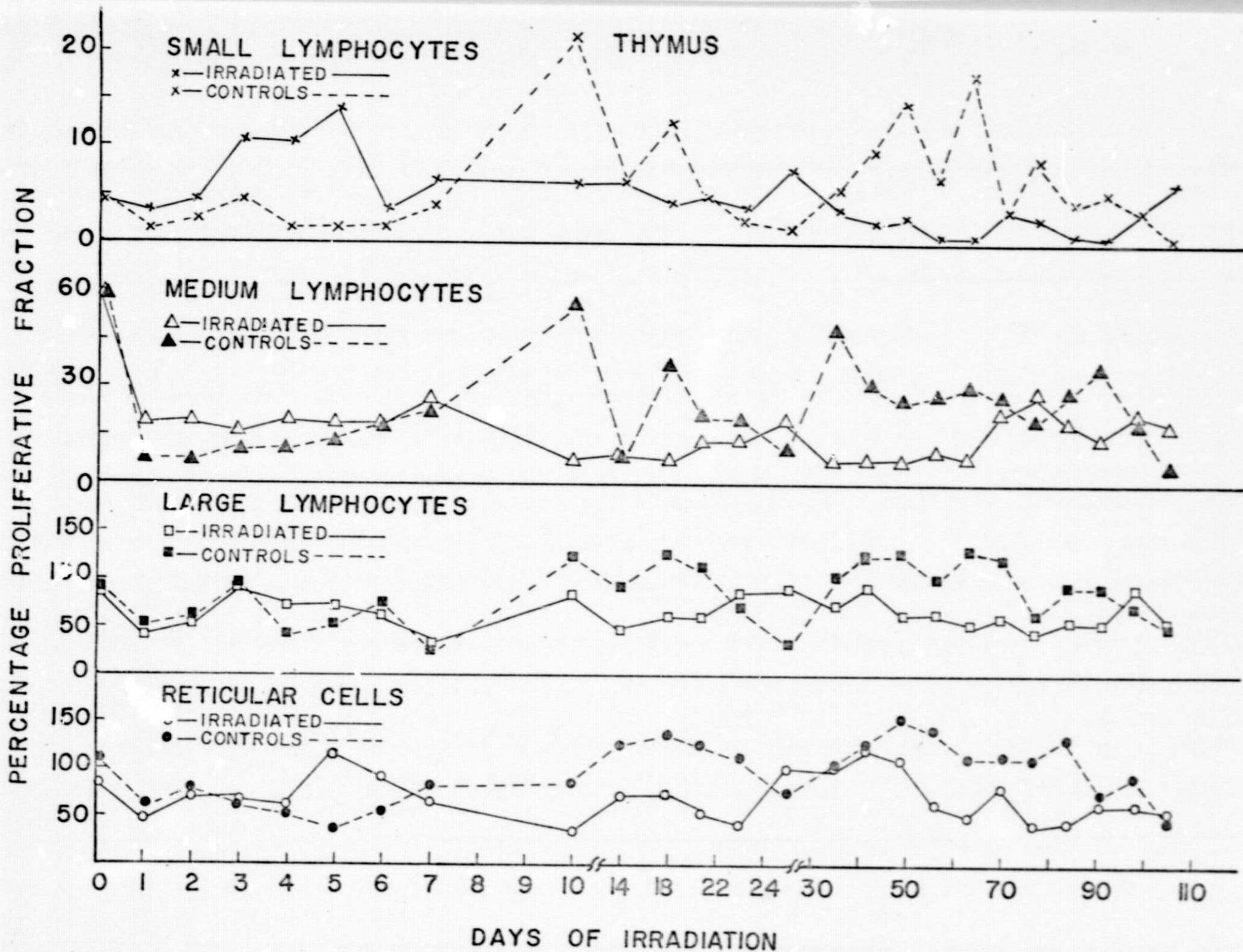


Figure 12

# SPLEEN

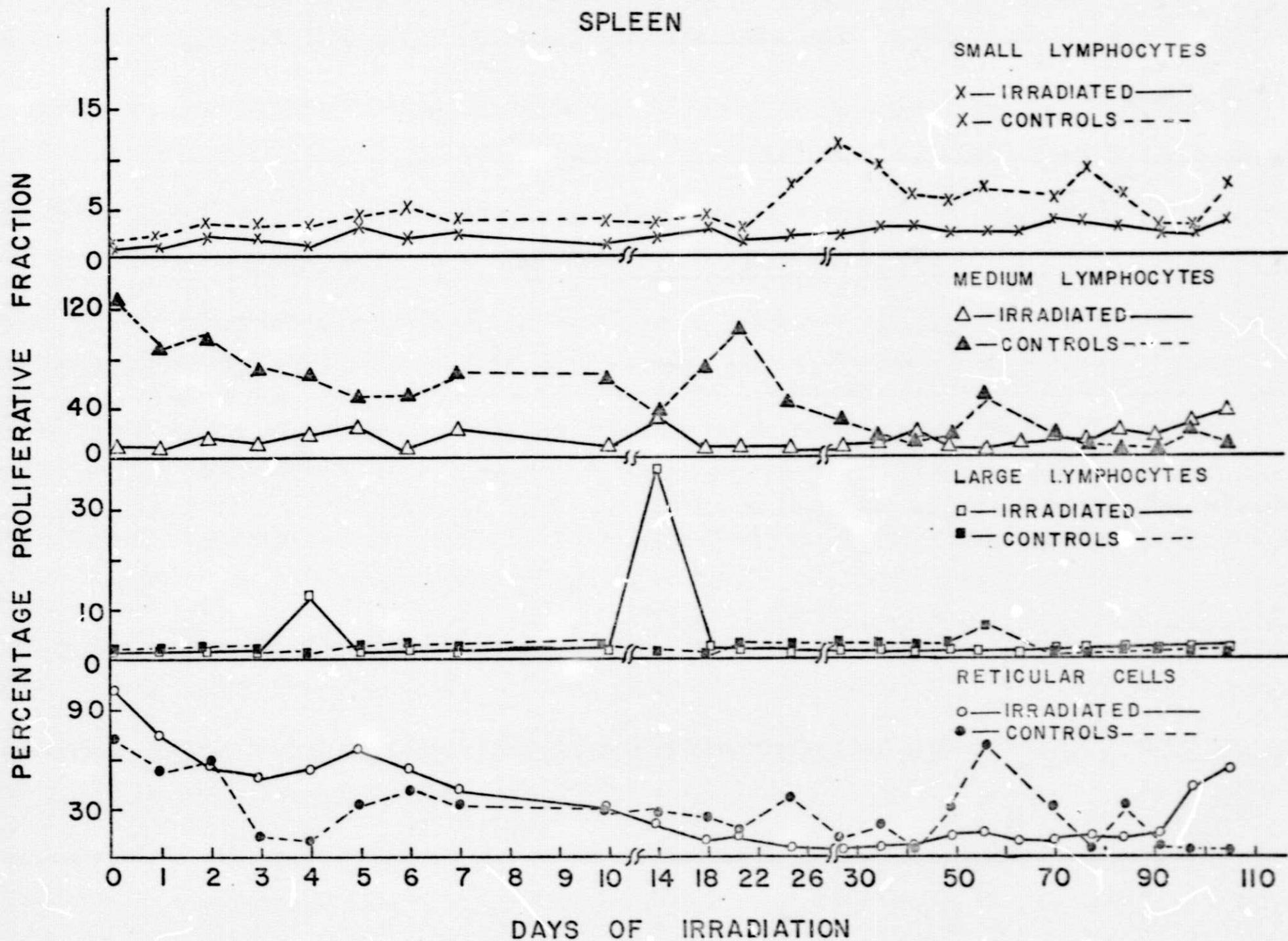


Figure 13

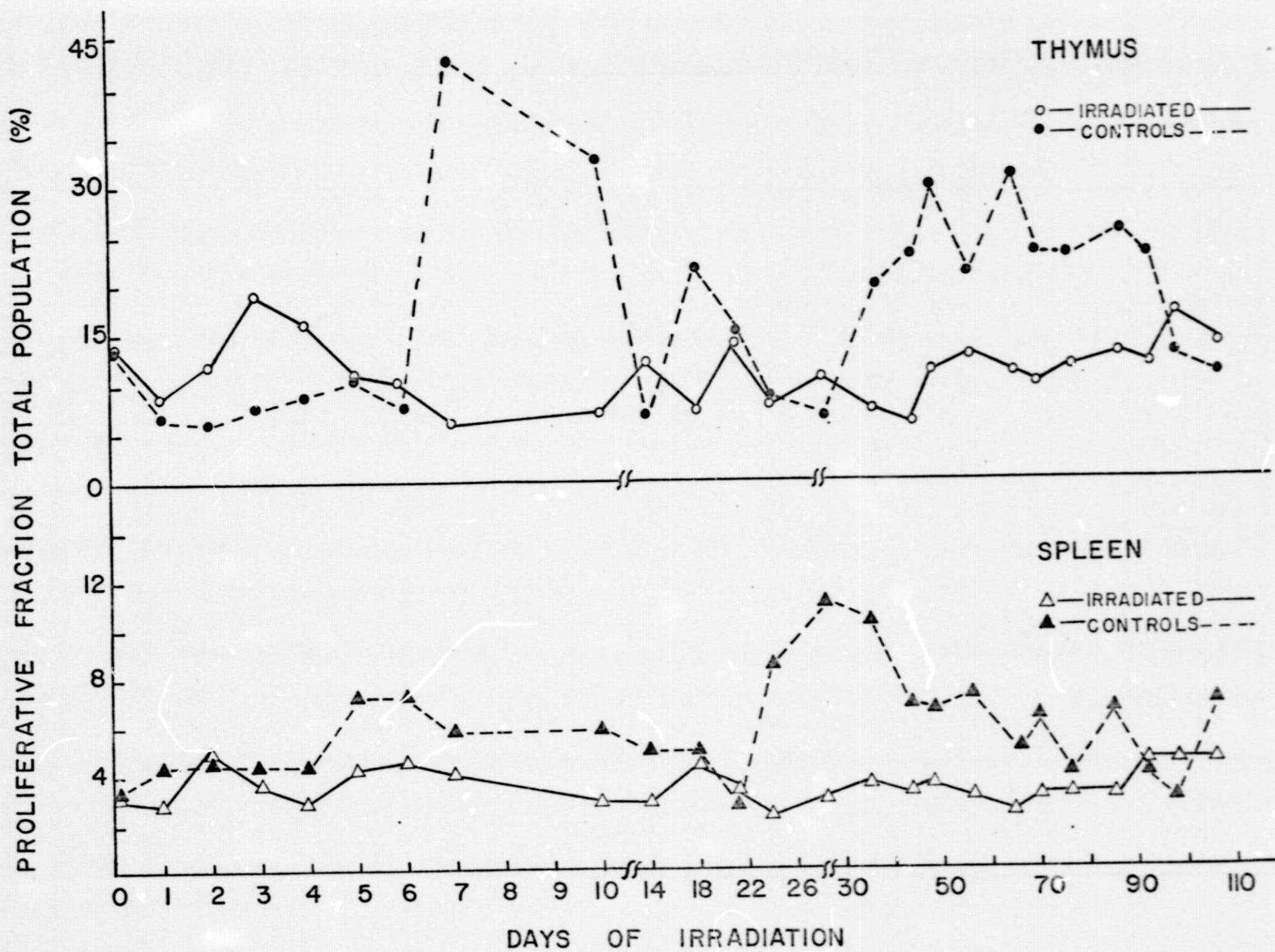


Figure 14

of irradiation, and an increase in the proportion of cells proliferating among all irradiated cell classes at the end of the radiation exposure. In the spleen, on the other hand, there was an increase in the proportion of irradiated reticular cells proliferating at the onset of the radiation treatment, and an increase in the proportion of cells proliferating among irradiated reticular cells and medium lymphocytes near the end of the radiation exposure.

Because of the relative numbers of medium and small lymphocytes, and because the small lymphocyte is thought to be rather non-dividing, the data further support the premise that the medium lymphocyte contributes most to the proliferative activity of lymphoid tissue.

Data on the proliferative fraction for the total population showed that there was an increased tendency for irradiated thymus and spleen tissue to proliferate near the end of the irradiation period.

The relative proliferative capacity was examined in lymphoid tissue in accordance with the method of Berman, Winter, and Newby (1966); by multiplying the percentage of each cell type times the percentage labeling in that category. These data are illustrated in Figures 15 and 16.

Among thymic cell types, there was generally an increase in the RPC among irradiated reticular cells compared to controls throughout most of the irradiation period. Also, there was an increase in the RPC among irradiated medium and small lymphocytes during the initial and final phase of the irradiation period. Most importantly, the medium lymphocyte exhibited an RPC of about 4 times that of reticular cells, and about 2 times that of large and small lymphocytes.

In the spleen, the RPC was greater among irradiated cells in the reticular and medium lymphocyte classes at or near the end of the radiation

# THYMUS

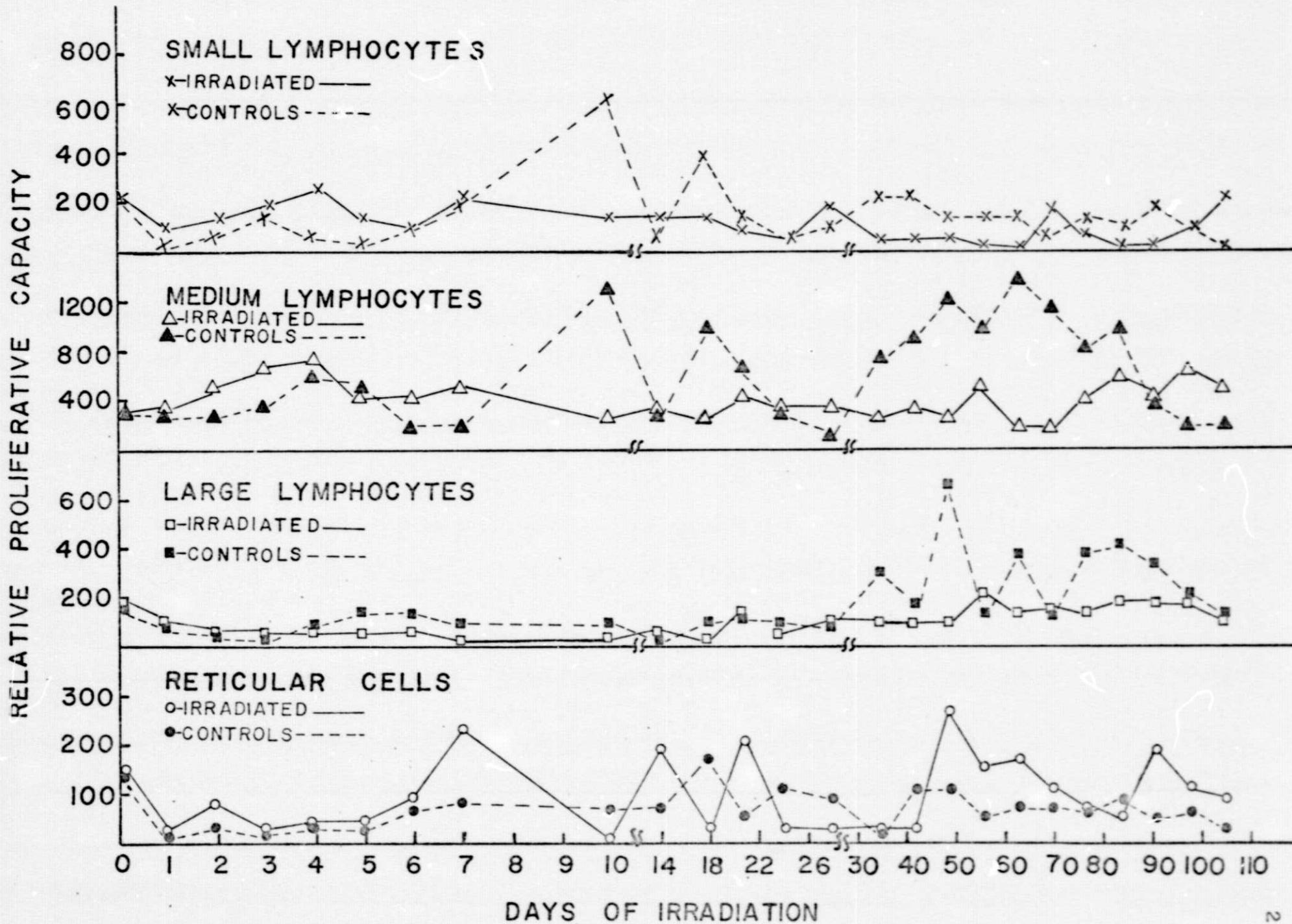


Figure 15

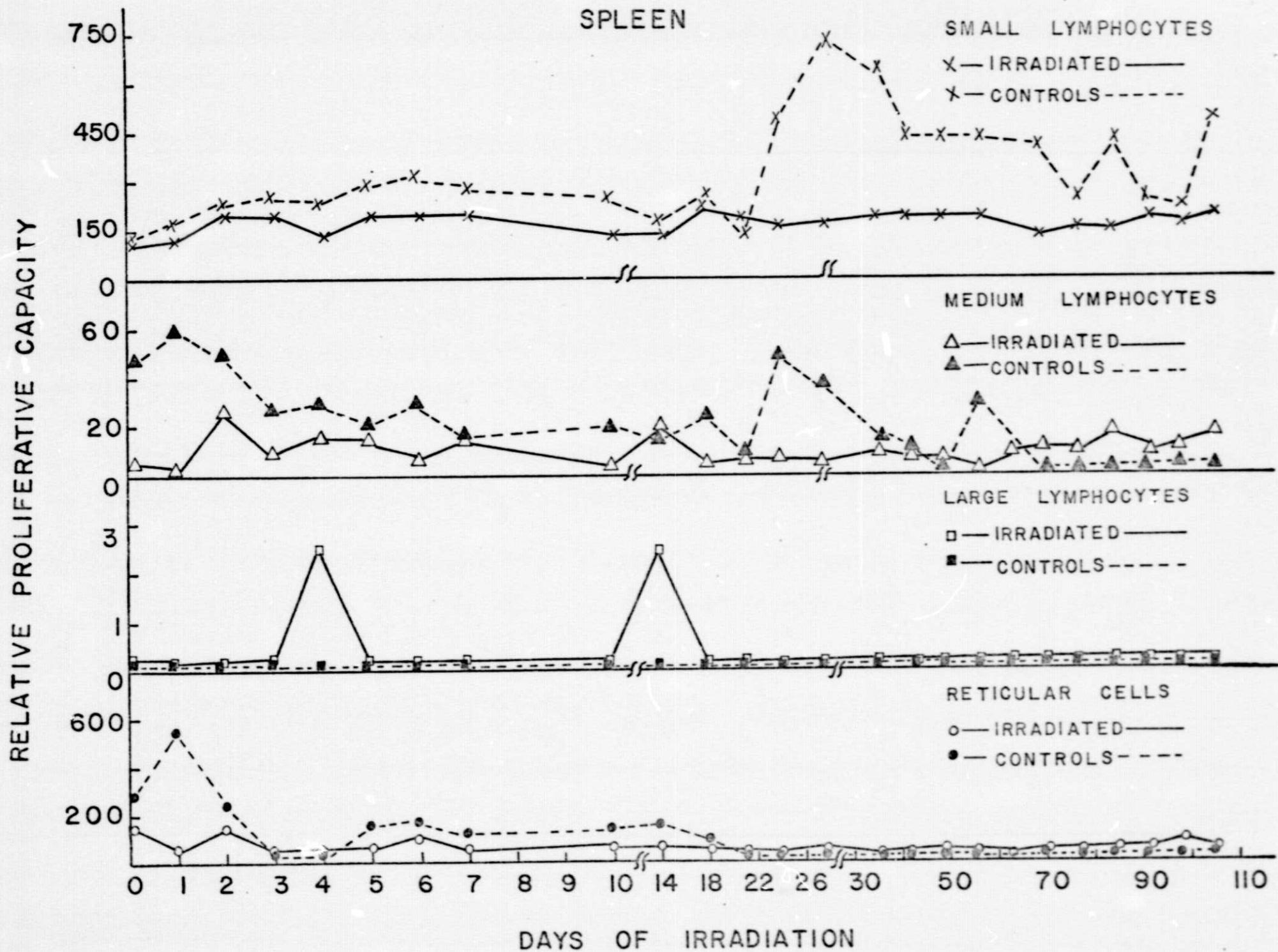


Figure 16

exposure. Again, it appears as though the medium lymphocyte, in particular, is highly proliferative among lymphoid cells.

Lastly, a comparison of findings observed in the thymus and spleen under continuous irradiation is found in Table 2. An average was taken on several parameters studied over the entire 105-day irradiation period. These averages suggest that there were about twice as many PAS-positive cells in irradiated thymus tissue compared to spleens. Among non-PAS reticular cells, labeling in non-PAS cells, and the proportion of lymphocytes; findings in thymus and spleen tissue were comparable. Labeling in thymus lymphocytes was about 6 to 7 times the values observed in spleens, labeling in the thymus population averaged about twice that observed in spleen tissue, and the maximum cell cycle time in thymus tissue was about one-half that calculated for spleen tissue.

#### Summary and Conclusions

On the basis of tissue weights, distribution of PAS-positive cells, cell cycle times, distribution of lymphocyte classes, thymidine labeling, proliferative fractions, and relative proliferative capacities of lymphoid cells under continuous irradiation, it was concluded that the response of both the thymus and spleen to a radiation dose of 10R per day for 105 days is seemingly bi-phasic. That is, a steady state or near-steady state of proliferation is achieved followed by tissue "collapse" followed by another steady state; the spleen being affected to the greatest extent.

Since there are two populations of lymphocytes--"short-lived" and "long-lived," it may be that the first steady state phenomenon is governed by the short-lived lymphocytes, and the second steady state phenomenon might possibly be mediated by long-lived lymphocytes.

TABLE 2

COMPARISON OF FINDINGS OBSERVED IN THE THYMUS AND SPLEEN UNDER CONTINUOUS IRRADIATION (AVERAGED OVER 105-DAY IRRADIATION PERIOD)

| Cell Type or Parameter                        | Thymus                               |            | Spleen                               |            |
|---|--------------------------------------|------------|--------------------------------------|------------|
|   | Percentage of Population or Category |            | Percentage of Population or Category |            |
|   | Controls                             | Irradiated | Controls                             | Irradiated |
| PAS Positive Reticular Cells                  | 0.11                                 | 0.12       | 0.00                                 | 0.05       |
| Non-PAS Positive Reticular Cells              | 4.22                                 | 4.06       | 5.89                                 | 4.54       |
| Labeling Non-PAS Positive Reticular Cells     | 37.01                                | 25.06      | 23.70                                | 20.16      |
| Lymphocytes                                   | 95.74                                | 95.22      | 94.00                                | 95.48      |
| Labeling Total Population                     | 10.00                                | 7.59       | 4.86                                 | 4.76       |
| Maximum Cell Cycle Time (Calculated) in Hours | 65.00                                | 85.60      | 136.50                               | 200.76     |

Compensatory mechanisms which enabled the radiation-depleted thymus cell population to achieve a steady state were:

- (1) an increase in the proportion of PAS-positive cells;
- (2) maturation arrest of precursor cells;
- (3) increase in the proportion of cells proliferating; and
- (4) an increase in the proportion of precursor cells.

Whereas, in the spleen, compensatory mechanisms which were operative were:

- (1) an increase in PAS-positive cells (only at onset of irradiation),
- (2) maturation arrest of precursor cells,
- (3) increase in the proportion of cells proliferating, and
- (4) an increase in the proportion of precursor cells.

Lastly, it was the medium lymphocyte which contributed most to the proliferative activity of the thymus, while the reticular cell class along with the medium lymphocyte seemed to have contributed most to the proliferative activity of the spleen.

## REFERENCES

- Berman, I., Winter, S.R., and Newby, E.J., (1966). *Anat Rec*, 154, 635.
- Brecher, G., Brambel, C.E., and Brambel, F.D. (1964). *Ann NY Acad Sci*, 114, 549.
- Cattaneo, W.M., Quastler, H., and Sherman, F.G. (1961). *Nature (London)*, 190, 923.
- Cronkite, E.P., Bond, V.P., Fliedner, T.M., and Rubini, J.R. (1959). *Lab Investigation*, 9, 263.
- Drew, R.M., and Painter, R.B. (1959). *Radiat Res*, 11, 535.
- Fabrikant, J.I. (1964). Ph.D. Thesis, University of London.
- Fabrikant, J.I. (1967a). *Radiat Res*, 31, 304.
- Fabrikant, J.I. (1967b). *Brit J Radiol*, 40, 487.
- Fabrikant, J.I. (1967c). *Radiat Res*, 32, 804.
- Fabrikant, J.I. (1968a). *J Cell Biol*, 36, 551.
- Fabrikant, J.I. (1968b). *American J Roentgenol Radium Therapy Nuc Med* 102, 811.
- Fry, R.J.M., Personal Communication.
- Gelfant, S. (1963). *Exp Cell Res* 32, 521.
- Goodwin, M.C. (1939). *Amer J Anat*, 64, 165.
- Gordon, A.S. (1955). *Ann NY Acad Sci*, 59, 907.

- Griem, M.L. (1966). *Nature (London)*, 210, 213.
- Hamilton, L.D. (1956). *Nature (London)*, 178, 597.
- Hamilton, L.D. (1958). *Ann NY Acad Sci*, 73, 39.
- Johnson, H.A. (1961). *Int J Cytol*, 26, 32.
- Johnson, H.A., and Cronkite, E.P. (1959). *Radiat Res*, 11, 825.
- Lala, P.K., and Patt, H.M. (1966). *Proc Nat Acad Sci*, 56, 1935.
- Lamerton, L.F. (1966). *Radiat Res*, 27, 119.
- Lamerton, L.F., Pontifex, A.H., Blackett, N.M., and Adams, K. (1960). *Brit J Radiol*, 33, 287.
- Leblond, C.P., and Sainte-Marie, G. (1960). *In Haemopoiesis*, 152, edited by Wolstenholme, G.E.W., and O'Conner, M., Boston: Little Brown.
- McCarter, J.A., and Quastler, H. (1962). *Nature (London)*, 194, 873.
- Mendelsohn, M.L. (1962). *J Nat Cancer Inst*, 28, 1015.
- Mendelsohn, M.L. (1963). *In Cell Proliferation*, 190 edited by Lamerton, L.F., and Fry, R.J.M., Oxford: Blackwell.
- Mendelsohn, M.L., Dohan, F.C., Jr., and Moore, H.A., Jr. (1960). *J Nat Cancer Inst*, 25, 477.
- Metcalf, D. (1964). *In the Thymus in Immunobiology*, 150, edited by Good, R.A., and Gabrielsen, A.E., New York: Harper & Row.
- Metcalf, D., and Ishidate, M., Jr. (1962). *Austral J Exp Biol*, 40, 57.
- Neary, G.J., Munson, R.J., and Mole, R.H. (1957). *Chronic Radiation Hazards*, London: Pergamon Press.

- Osgood, E.E. (1957). J Nat Cancer Inst, 18, 155.
- Osgood, E.E. (1961). Ann NY Acad Sci, 95, 828.
- Ottesen, J. (1954). Acta Physiol Scand, 32, 75.
- Painter, R.B., Drew, R.M., and Hughes, W.L. (1958). Science, 127, 1244.
- Post, J., and Hoffman, J. (1961). Radiat Res, 14, 713.
- Quastler, H., and Sherman, F.C. (1959). Exp Cell Res, 17, 420.
- Sainte-Marie, G., and Leblond, C.P. (1958). Proc Soc Exp Biol Med, 98, 909.
- Steel, C.G., Adams, K., and Barrett, J.C. (1966). Brit J Cancer, 20, 784.
- Trowell, O.A. (1952). J Pathol Bacteriol, 64, 687.
- Trowell, O.A. (1961). Int J Radiat Biol, 4, 163.
- Vos, O. (1967). In Effects of Ionizing Radiations on Haematopoietic Tissue, 134, Proceedings of a Panel. Vienna: IAEA.
- Wimber, D.E. (1959). Proc Nat Acad Sci, 45, 839.
- Wimber, D.E. (1964). In Advances in Radiation Biology, 85, edited by Augenstein, L.G., Mason, R., and Quastler, H., New York: Academic Press.
- Yoffey, J.M. (1960). Quantitative Cellular Haematology, edited by Bethell, F.H., Springfield, ILL: Charles C. Thomas.
- Yoffey, J.M., Reinhardt, N.O., and Everett, N.B. (1961). J Anat, 95, 293.

## PUBLICATIONS, THESES, INVENTIONS AND DISCOVERIES, AND SCIENTIFIC COLLABORATORS

Publications

The following publications have been made pertaining to the research (associated with NASA Grant NGR-19-011-008 and NSF Grant GB-29136), copies of which have been forwarded to NASA Scientific and Technical Information Facility.

- (1) Foster, B.R.. The Effect of Continuous Low Dose-Rate Gamma Irradiation on Tissue and Body Growth. The Louisiana Physics Teacher. Volume 3, Number One, Spring, 1973, pp. 17-19.
- (2) Foster, B.R., Taylor, B., LeFear, P. and Wake, M. Biphasic Response of Lymphoid Tissue Under Continuous Irradiation. Radiation Research. Volume 59, Number 1, July, 1974, p. 304 (Abstract).

At least one other publication will be made in the form of a research paper, associated with NASA Grant NSG 9014, to be published in journals such as Radiation Research, the manuscript of which is in preparation. When the report is published, reprints will be forwarded to NASA.

Theses

No thesis was prepared in connection with the research.

Inventions or Discoveries

No inventions nor discoveries were made in connection with the research.

Scientific Collaborators

The scientific collaborators connected with the grants include one senior scientist, and a total of 23 pre-baccalaureate students who studied and worked with the project at some given time. The senior scientist is

Dr. R.J.M. Fry of The Argonne National Laboratory, Argonne, Illinois, who served as my advisor during the summer, 1971, when the principal investigator was a participant in the Argonne Faculty Research Participation Program. During this time (summer, 1971), the initial phase of the study, that is, the irradiation of the test animals, was carried out in the laboratory of Dr. Fry.

The following pre-baccalaureate students studied and worked with the project:

Billy Taylor - physics major (graduated)  
 Joseph Alexander - physics major (graduated)  
 Joseph Blow - mathematics major (graduated)  
 Neda Bailey - mathematics major (graduated)  
 Ronnie Blake - chemistry major (senior)  
 William Wiley, III - physics major (senior)  
 Alan Kennedy - chemistry-computer science major (senior)  
 Matthew Ware - physics major (graduated)  
 Caffin Gordon - sociology major (graduated)  
 Dorothy Baker - business major (graduated)  
 Sharon Harris - pre-medicine major (senior)  
 Patricia LeFear - physics major (senior)  
 Don Blow - mathematics major (senior)  
 Cynthia Tizeno - physics major (junior)  
 Michael Banks - political science major (junior)  
 Larry Nicks - chemistry major (junior)  
 Gerry Mansfield - physics major (sophomore)  
 Dennis Dowell - physics major (no classification)  
 Angelia Young - biology major (freshman)  
 Gregory Route - recreation major (graduated)  
 Malloy Sanders - special student (unclassified)  
 Gilda Harris - special student (unclassified)  
 Debra Handy - social work major (senior)

#### Comments

Other than making possible the development of a well-equipped research laboratory and actively engaging in the research project per se, the greatest contribution that the research grants made to the University was the involvement of undergraduate students in research procedures. Students became familiar with routine laboratory and research procedures,

learned how to manipulate research equipment, and became adept in collecting and interpreting data. Research experiences which the students realized will be beneficial to them in graduate school, industry, or whatever career they choose to pursue.

The funds allocated for student support enabled some students to earn their college tuition or other fees, many of whom would not have been able to pursue a college career otherwise. I personally hope that NASA will continue to look favorably upon research proposals that actively involve undergraduate students at developing institutions.