# RECORD KEEPING FOR LABORATORY ANIMALS

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(With 6 Figures in the Text)

The amount of clerical work involved, and the ramifications of the record system used, in an animal laboratory, are conditioned by the purpose for which the colony is maintained, and dependent upon how anxious its custodians are to control its size and guide its future. If, for instance, ease of operation and minimization of clerking is to be the overriding consideration one may secure a good supply of young rats by the simple process of allowing a flock of adult animals the freedom of a warm room containing bedding and food. One enters at intervals to clean and feed, and, eventually, to remove, for experimental work, those youngsters that look big enough to fend for themselves, taking care, at the same time, to leave a few of each sex to grow to maturity and guarantee the next generation. An occasional census of population and production is all that is necessary in the way of records.

This ménage has actually been used, although with what success, and for how long, I cannot say. Its advantages—apart from providing material for a study of social behaviour in the animal world—would seem to be economical, in several obvious directions. Yet it would in all likelihood prove in the long run to be false economy, in so far as: (*a*) the output of young would be spasmodic, difficult to anticipate, and almost impossible to control; (*b*) any undesirable characteristics manifested could not be genealogically 'taped' and hence restricted or eliminated—similarly, good characteristics, such as weight and litter size, could not be selectively bred; and (*c*) the number of young required per experiment would in most cases be greater, for a given degree of precision, than that from a controlled colony. In other words, the measurable characteristics of rats bred at hazard and subsequently randomized usually display a greater variance than those of selectively bred and litter-segregated rats.

A controlled colony may be defined as one whose breeding is planned and whose genealogy is the ever-present concern of its directors. Most modern colonies are of this type. They obviously necessitate some fairly elaborate system of record keeping. I propose to describe two such systems, both applicable to rat colonies.

Any discussion of written records needs to be prefaced by a brief account of how the animals themselves are marked for identification purposes. A simple and wholly satisfactory marking device for small animals is not easily found, yet a faulty device may render the work of the most careful recorder all but useless.

The method used in the writer's laboratory is ear marking, perhaps the commonest and indubitably the simplest. Chicken-toe punches make circular holes, about $\frac{1}{16}$ in. diameter, and the pinna of the ear of a weanling rat will take three of these holes. As there are two ears and four different holing possibilities in each (0, 1, 2 or 3 holes) the total number of individuals so identifiable is 16. This means, in effect, that identification is limited to one or two litters, or one or two large cagefuls of rats, and particular care has therefore to be taken to prevent straying. The number of identifications can be raised to 32 if an extra hole per pinna is allowed, but we find that the unavoidably frail scissel left in a 4-punched ear is liable to tear and to grow unrecognizable. Some American laboratories combine ear marking and toe clipping—amputation of the small toe at the first joint. In this way a total of $16 \times 16$, or 256, individuals can be identified. Toe clipping is, however, illegal in Great Britain. Another American device, due to Keeler (1940), is a tattoo punch that can prick any of the ten numerals into the pinnae, the pricks being subsequently Indian inked. Two numerals per ear will distinguish 10,000 animals; three will cover 1,000,000. A third method is to mark the tails with a dye such as basic carbol fuchsin stain, but in our experience the markings tend to fade and wear, and identification becomes difficult.

Given a suitable marking scheme, we now turn to the clerical side, and begin with a consideration of what to record. The following are suggested as essential data: (*a*) pedigree, (*b*) birth, weaning and mating calendar, (*c*) litter sizes, sex distribution and growth rate, and (*d*) infant mortality. Additional data depend on the trouble the recorder is prepared to take, and the strength of his feeling that they may 'come in useful'.

The scheme used at the Wistar Institute, Philadelphia, the *fons et origo* of all rat colonies, is wholly card recording (Griffith & Farris, 1942). The breeding

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practice is to allow three does and one buck to cohabit for 5 days, to segregate the does immediately after mating, to rest the mothers for the 2 weeks following the birth of a litter, and to restrict littering to a maximum of four per mother. Each rat is given a kind of escutcheon carrying all the information necessary to determine its position on the family tree. An example is shown in Fig. 1. Each breeding

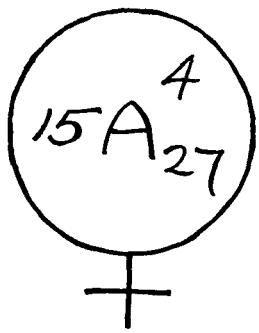


Fig. 1. Wistar escutcheon. Serial letter (A), series or line; index number (15), generation; superscript (4), whether from 1st, 2nd, 3rd or 4th litter of mother; subscript (27), chronological number of the litter among, in this case, all the 4th litters of the 15th generation.

this availability will vary according to the management of each laboratory. In essence the scheme consists of the permanent housing together of an adult litter (i.e. from about 100–120 days of age) containing a predominance of does, for example 6 does and 2 bucks. The does are inspected for pregnancy twice weekly and, when obviously within 4 or 5 days of the end of term, are removed and placed in individual littering cages. There the litters are cast and nursed for 23 days—or a little longer if the members of the litter average less than 30 g. After weaning the mother is immediately returned to the family mating cage, i.e. no rest period is given. If littering performance is good, up to 6 or 7 pregnancies are allowed. Although 6th and 7th litters rarely make good breeders themselves, they are perfectly satisfactory for experimental work. Most of the weanling litters are in fact taken for experimental purposes, but a minority, comprising about 5 % of the whole, is 'put back into stock', i.e. raised to become breeders. The selection of these stock litters is based on sex ratio and pedigree.

This regimen does not take account of mating dates, nor does it permit identification of the father among the 2 or 3 bucks in the breeding litter. Furthermore, records are not kept of the number, sex or weight of newborn young. None of these data

Cage 409

Female 1 A 5 52 ♂ Born 1-5-39 Parents 4C × 3C

Diet

| Male | Date | Wt. | Wt. wks. later | Date litter born | Born dead | ♂ | ♀ | Wt. litter | Fed | Date weaned | ♂ | ♀ | Wt weaning | Litter no. |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 A 2 45 ♂ | 5/8 | 178 | 251 | 6/9 | | 6 | 4 | 62 | ✓ | 6/29 | 6 | 4 | 313 | 2A' 271-278 |
| | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | |

Fig. 2. Wistar breeding card (see Griffith & Farris, 1942). Reproduced from *The Rat in Laboratory Investigations* (p. 14), by kind permission of the authors, J. Q. Griffith and E. J. Farris, and the publishers, The J. B. Lippincott Company.

cage has a card bearing the escutcheon, the dates of birth and mating, and the cage number. Cages for weanling rats bear cards giving the litter number, the number and sex of the individuals, the date of birth, and the escutcheons of both parents. Finally, each female breeder has a large filing card giving the data shown in Fig. 2.

The scheme obtaining in the writer's laboratory is simpler than that of the Wistar Institute, the simplicity being mainly at the expense of multiple availability of pedigree details. The importance of is important to us (although they may be in other laboratories) and ignorance of them in no way limits assessment of breeding performance, as all birth statistics are reflected in weaning statistics.

Let us now suppose that a certain doe, number 7667, in cage 114, casts a litter, her third, on 1 February. Entries are thereupon made in two loose-leaf books, which together contain all the basic records, no other books or cards being kept at this level. The first of these books is the Birth Register; the sheets have the headings shown in Fig. 3,

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where the new entry is given. The serial number of the newborn litter (first column) follows on from the preceding entry. The final item in the line, the composition of the litter, will not be entered until weaning time. Each sheet in the book has space for 100 litters, and a new sheet is opened at the beginning of each calendar month. When a month's entries are complete, the numbers of bucks and does weaned, and of litters born, and the litter mortalities during nursing, are totalled. It may be noticed that it is to this register that the staff turn every morning to ascertain what weanings are due; it thus obviates the keeping of a diary.

In the second loose-leaf book, the Breeding Stock Book, each sheet covers the record of one breeding litter. The sheets are arranged chronologically, one side providing for adolescent growth data, the other for breeding details. So we now turn up the litter of which number 7667 is a member and find that the last entry for this doe records her segregation, from her litter mates, on 27 January, when she was observed to be in an advanced state of pregnancy. The appropriate further entry is now made and Fig. 4 shows the state of the sheet. The final job in connexion with the event is to pencil on the cage ticket (allocated on 27 January with the number and ear markings of the mother) the date of birth and the serial number of the newly cast litter. (Cage tickets are of blank cardboard, $4 \times \frac{3}{4}$ in., that slide into metal slots below the cages.)

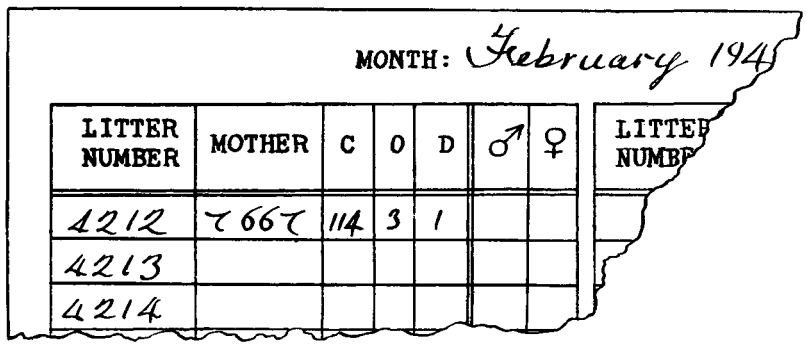


Fig. 3. Section of loose-leaf of Birth Register. C, cage number; O, order of litter; D, date (day of month only) of birth; ♂, ♀, composition of litter when weaned.

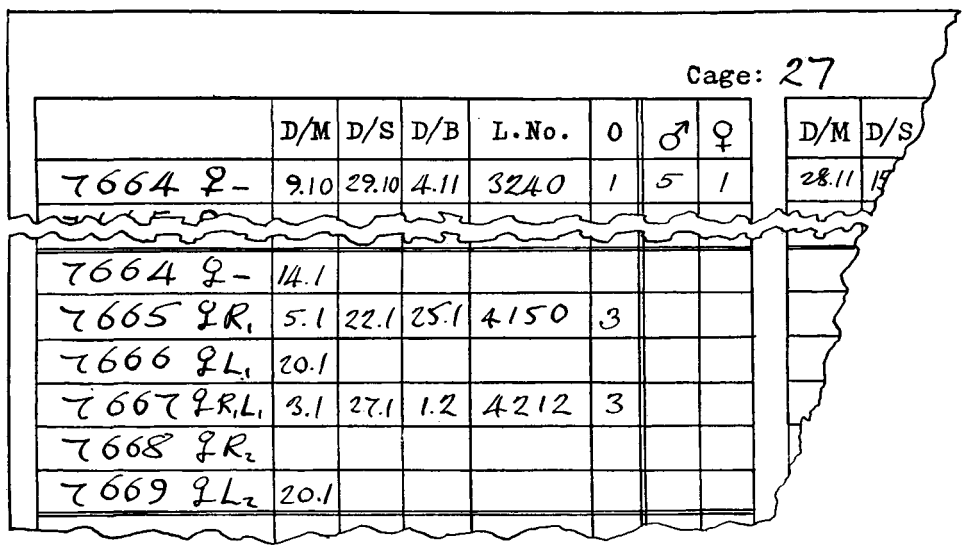


Fig. 4. Section of loose-leaf (verso) of Breeding Stock Book. D/M, date of mating; D/S, date of isolation of mother; D/B, date of birth of litter; L.No., serial number of litter; O, order of litter; ♂, ♀, composition of litter at weaning.

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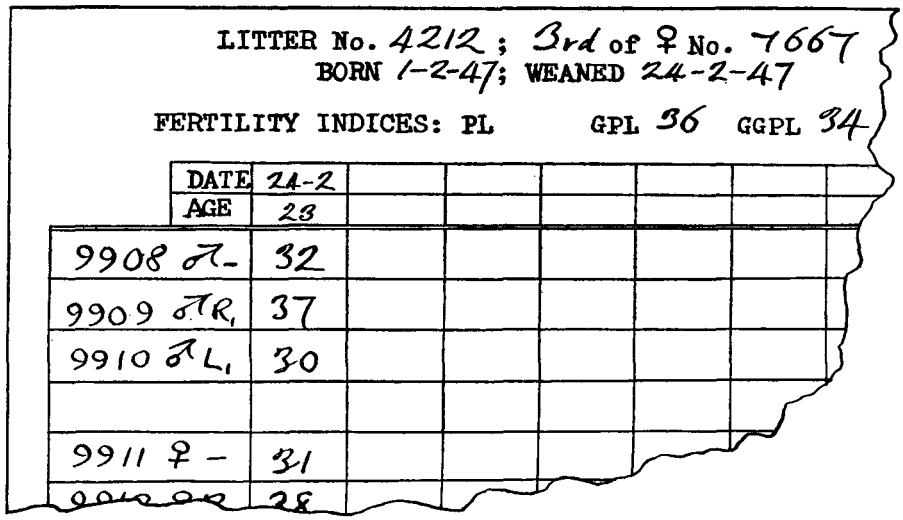
Provided that the litter survives the nursing period, no further clerical work is done until 24 February, when the litter is due to be weaned. Let us assume that on that date the litter is found to contain 3 bucks and 7 does, all of normal size. This is a convenient sex ratio for a breeding litter, so we look at the 'fertility index' (about which, *vide infra*) distributed in time, and, *ceteris paribus*, between and within families.

Our newly weaned stock litter is weighed, ear-marked and given the next ten numbers in a series that embraces all weanlings chronologically, no matter what future is intended for them. A fresh sheet in the Breeding Stock Book is now opened.


LITTER No. 4212 ; 3rd of ♀ No. 7667
BORN 1-2-47; WEANED 24-2-47
FERTILITY INDICES: PL GPL 36 GGPL 34

| DATE | 24-2 |
|---|---|
| AGE | 23 |
| 9908 ♂ - | 32 |
| 9909 ♂ R, | 37 |
| 9910 ♂ L, | 30 |
| | |
| 9911 ♀ - | 31 |
| [illegible] | 28 |

Fig. 5. Section of loose-leaf (recto) of Breeding Stock Book. Main entries are weights in grams; PL, GPL, etc., parent litter, grandparent litter, etc.

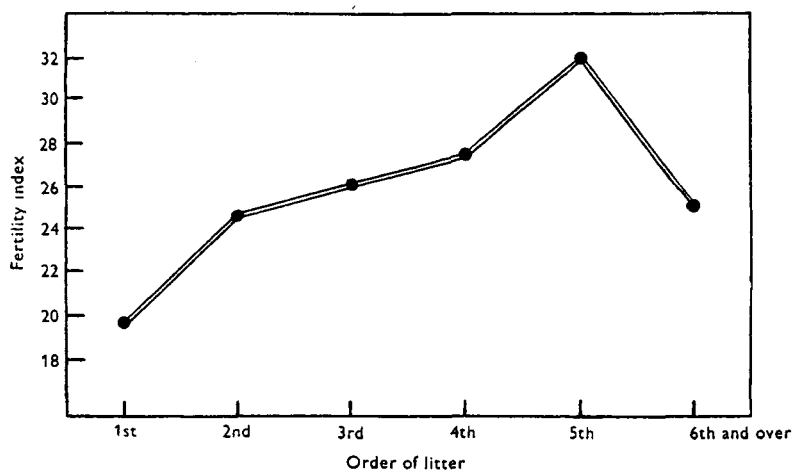



Fig. 6. Curve showing variation of fertility with litter order (and, in effect, with age of parents at birth).

and as it is high, we decide to 'take the litter into stock'. The entries in the two loose-leaf books (Figs. 3 and 4) are therefore completed with the figures 3 and 7 in red ink. As ordinary blue ink is used for the entering of weanlings taken for experimental work (i.e. the majority), the colour contrast helps the staff to see that the intake of fresh stock is evenly

Fig. 5 shows the layout (the other side has the layout of Fig. 4, but as yet blank), and the serial numbers, ear markings and weaning weights are entered as there shown. The rats are subsequently weighed at intervals of not less than 14 days, the bucks and does put into separate cages at about the 50th day of age, and reassembled (i.e. mated) somewhere between the

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105th and 120th days. The other side of the sheet is then brought into use, and the breeding record begun.

This method of recording is thus analogous to double-entry book-keeping. It makes for easy accessibility of essential information from two points of view, chronological and genealogical. And the keeping together of the breeding record of a sisterhood of does, in contrast to the more usual one-card-per-breeder system, ensures continual surveillance of family performance. From time to time completed pages of both books are removed and a digest of their contents put into a large notebook. This contains tables showing the month-by-month, seasonal and year-by-year history of the colony in terms of birth-rate, sex ratio and infant mortality (i.e. percentage litters born that are killed by their mothers or otherwise do not survive the nursing period). It also contains serial records of the genealogy and performance of all breeding families, the most important criterion of performance being what we call a 'fertility index', defined as 'mean number of weaned offspring per breeding doe excluding seventh (and over) pregnancies'. The average index is about 25; the best families show more than 50. It is an extremely useful criterion to base breeder selection on. The indices of the immediate progenitors are put on each breeding sheet, as shown in Fig. 5.

The ultimate condensation of the records takes the form of progressive charts to show at a glance the present state and past history of the colony. We chart such statistics as monthly and yearly production, and plot fertility indices against time, generation and litter order. Fig. 6 is an example of the last-named.

I end with an illustration of the many useful purposes served by good, and adequately condensed, records. Rats are prolific animals, an average doe producing, under the breeding regime of the writer's laboratory, 25 or so offspring in her useful life-span of 12 months. This means that to maintain the colony at a given size, in other words to stabilize the production of experimental material at a chosen rate, it is necessary to 'put back into stock' approximately one doe per 25 weanlings. This fact, together with the curve of the seasonal variation in fecundity, provides a basis for a decision on the best rate of stock replenishment from month to month throughout the year to ensure a more-or-less constant rate of production of experimental material in the future. And a programmed increased or decreased rate of production can be met within narrow limits of error. Furthermore, if, on top of this replacement-ratio datum, we take into account—into quantitative account, that is—such statistics as the average size and sex ratio of a breeding litter, and the total accommodation required for breeding, nursing and maturing rats, we can establish a relation between production and accommodation (Table 1). The maximum capacity of the existing laboratory space and equipment is then clear-cut, and the expansion required for any required increase beyond this maximum can be given with confidence.

Table 1. *Relation between breeding rate and accommodation*

| Breeding batteries available | Average young per month | Littering batteries required | Minimum floor space (sq.ft.) |
|---|---|---|---|
| 1 | 155 | 2 | 93 |
| 2 | 325 | 4 | 186 |
| 4 | 685 | 7 | 341 |
| 8 | 1405 | 13 | 651 |
| 12 | 2135 | 21 | 1023 |

N.B. A 'breeding battery' consists of twelve large cages, a 'littering battery', twenty-four medium cages.

## REFERENCES

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