counts of 1,374 grains gave 85 per cent. of normal-appearing grains, whereas only .5 per cent. were good in a sample of 1,465 grains of F_1 pollen. The doubling has evidently resulted in the establishment of an amphidiploid condition, as is to be expected in sterile hybrids of this kind.

The hetero-auxin treatment has also been applied successfully to plants growing in the field. Under these conditions plants of *N. glauca, N. tomentosa, N. tomentosiformis* and F_1 *sylvestris-tomentosa* have produced callus shoots abundantly and tetraploid shoots have been found among them in even higher percentages than in the greenhouse. Plants of *N. tabacum* (Maryland Mammoth, *purpurea* and *purpurea Mammoth*), F_1 *glutinosa-tomentosa* and F_1 *glutinosa-sylvestris* have produced a few callus shoots, among them some tetraploids. Plants of *N. rustica* (*brasilia*), *N. wigandioides* and F_1 *glutinosa-tomentosiformis* have produced callus tissue and roots but as yet no callus shoots. It seems probable, however, that the relatively poor reponse in these instances is to be ascribed to poor condition of the plants, rather than to unsuitability of the treatment. To be regularly successful, the treatment must be applied to young plants in good growing condition.

Successful outcome of the treatment appears to depend upon careful attention to a number of details. Plants should be decapitated so that five or more good leaves remain on the plant. Internodal cuts respond to treatment just as well as those made through or close to nodes. The cut surface should be covered with a thin layer of the hetero-auxin paste immediately after decapitation, after which it should be protected, especially if exposed to direct sunlight, by covering with two or three folds of muslin or with a manila paper bag loosely tied to the stem. When buds begin to develop the covering should be gradually removed. It is important to have the plants in good growing condition and to keep them so during the course of the experiments. The plants should be disbudded immediately after treatment and any new axillary shoots should be removed as they appear. Successful treatment is followed by prompt development of callus tissue, which may in some instances take on a rounded, tumor-like appearance. After four to six weeks buds begin to develop, often so thickly as to completely cover the callus surface with a close tuft of shoots. The shoots may be classified when two or three inches long by examination of strips of epidermis peeled from the lower surface of the leaves. Callus shoots may be removed and treated as cuttings, or one or two selected ones may be left to develop in place after removal of the rest. There seems to be a tendency for normal shoots to develop more promptly than polyploid ones, so that it is advisable to remove the earlier shoots as soon as they are large enough to be examined, in order to give the less rapidly developing polyploid shoots a chance.

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ADENOMATOUS LESION IN STOMACH OF STRAIN I MICE

THE I strain of mice was originated by Dr. L. C. Strong[1] in 1927 by crossing a pink-eyed dilute brown and a dilute brown piebald stock. From this cross he selected five recessive characters, namely: pink-eyed, dilute, brown, non-agouti and piebald and continued the strain by brother-to-sister mating. Female mice of this strain seldom lived longer than fourteen months.

A litter of strain I mice was procured from the Roscoe B. Jackson Memorial Laboratory and the progeny of these mice, obtained by brother-to-sister mating, have been used as experimental animals in this laboratory. The strain has been of considerable interest for, up to the present time, none of the breeding females has developed a spontaneous mammary gland carcinoma. Furthermore, the mice have proved to be very resistant to the growth of transplantable mouse sarcomas 37 and 180 as well as resistant to the induction of subcutaneous tumors by lard solutions of carcinogenic hydrocarbons.

It was during the course of an experiment in which strain I mice had been injected subcutaneously with a lard solution of 1:2:5:6-dibenzanthracene that the stomach lesion was first encountered. The question arose as to whether the lesion was induced by the carcinogenic compound or whether it occurred spontaneously in members of this strain. Consequently strain I mice which had not been subjected to any experimental procedure were sacrificed and examined for the presence of stomach growths.

Twenty-one mice, aged 2.5 to 15 months, were killed and autopsied immediately. At three or four months, a progressive gastric lesion began, which was fairly comparable in brothers and sisters and in more distant relations of the same age. Practically all animals ten months old and over showed the condition in advanced form.

The principal change occurred in the pyloric chamber of the glandular portion of the stomach, which became large, solid, firm, opaque and slightly congested, with small grey nodular elevations on the serosa. In old mice the stomach contained little food and the cardiac chamber was compressed against the

[1] This information concerning strain I mice was kindly supplied by Dr. Strong in a personal communication.

costal cage. The glandular mucous membrane was thickened with coarse, grey, hypertrophied rugae. This change began abruptly at the constriction in the middle of the stomach and decreased progressively, disappearing near the pyloric orifice, with the most marked involvement along the greater curvature. Microscopically, the changes consisted of a pronounced hyperplastic adenomatous overgrowth of the glandular mucous membrane, with coarse polypoid projections resembling hypertrophied rugae. The glandular structures became dilated and irregular in outline with the formation of microcysts in some specimens. Collections of inflammatory cells were scattered through all coats of the viscus. The cardiac chamber only occasionally showed slight hypertrophy and hyperkeratosis of the squamous epithelial lining. Coincident with the development of the lesion, the normal distribution of the various cell types in the glandular mucous membrane was disarranged. The epithelial cells became irregular in size, shape and staining, and showed squamous, cuboidal, polygonal and columnar forms with numerous mitotic figures. Some cells were necrotic, desquamated and keratinized and a few acini appeared completely filled with keratin. Although the process usually appeared superficial and well limited to the mucosa, in several specimens distended acini were observed indenting the basement membrane and lying partly below it. Furthermore, in nine of twenty-one cases, a few small fragments of atypical epithelial cells were found below the basement membrane, usually in the submucosa but also in the muscle and, in two cases, in the lumens of veins. These deeply situated deposits rarely consisted of more than one or two fragments in a given specimen and their significance in this connection has not been definitely established. No metastases were observed in any instance. Starvation, dependent upon alteration of the mucosa and occlusion of the gastric lumen appeared to be the cause of death in many mice of this strain.

So far, with the exception of three old strain C_3H mice aged 22.5, 23 and 24 months, analogous lesions have not been encountered in the stomachs of mice of various other pure strains.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

AN IMPROVED INSTRUMENT FOR THE INTRACEREBRAL INOCULATION OF EXPERIMENTAL ANIMALS

THE inoculation of animals by the intracerebral route is a procedure frequently employed in research and diagnostic laboratories. This method necessitates the use of a pointed instrument to penetrate the skull, except when the experimental animals are young mice. Of the wide variety of commercially available trephines and improvised instruments now commonly used for this purpose in different laboratories, we have not found any that is as satisfactory as the instrument to be described in this note. Because of its construction, it can not be inserted too deeply, thereby injuring the brain. Furthermore, it has the advantage of a groove which permits insertion of the inoculating needle without necessitating the withdrawal of the instrument, thereby eliminating the possibility of losing the hole through the skull. Any one who has used the intracerebral method for inoculation has experienced this time-consuming annoyance.

The instrument may be made of any suitable material. We have found that common nails serve admirably. They are cheap and universally available. A small vise and two steel files are all the equipment required. The procedure is as follows:

(1) File off one side of the nail to make an oblique plane (Fig. 1–A).

(2) Using a three-cornered file, make a longitudinal groove down the middle of the oblique plane (Fig. 1–B).

(3) File a circular shelf near the point, extending inward for one third of the nail's diameter (Fig. 1–C).

(4) File a triangular point, the size and depth of which is determined by the thickness of the skull of

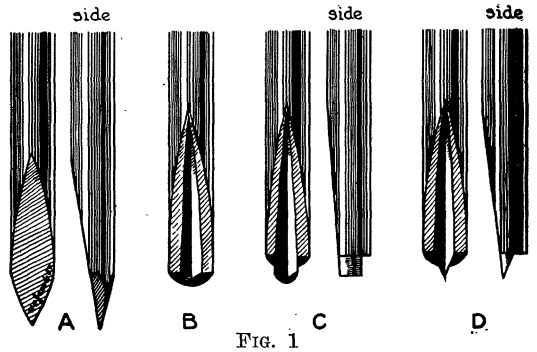

FIG. 1

the experimental animal for which the instrument is destined. This point is separated from the rest of the instrument by the shelf (Fig. 1–D), which serves to prevent penetration of the skull more deeply than desired (Fig. 1–D). By extending the groove (Fig. 1–A) to the very point, the inoculating needle can enter the brain substance without necessitating withdrawal of the instrument.