especially twins and triplets (Fig. 2, Nos. 3, 10 and 12), throw normal as well as abnormal individuals. Some of the abnormal forms, however (Fig. 2, Nos. 7, 11, 15 and 16), breed true and have been kept in pure culture for many generations by sub-culturing with a platinum loop. Thus, long exposure of Paramecium to benzpyrene results in the production of a set of polymorphic cells. Here we have a striking resemblance to the assemblages of cells of which tumours are composed; it is well known that the cells from a single tumour are widely polymorphic both in size and structure: for example, their content of chromosomes may be either normal, above the normal, or below; cells with many times the normal number are common.

The similarity between the formation of a tumour by benzpyrene and the changes observed in Paramecium thus embraces the following facts:

(1) They occur among cells long stimulated to a growth-rate above the normal.

(2) They occur only after long exposure to the hydrocarbon.

(3) Only a very few of the exposed cells present the change.

(4) Once the change has occurred, it is reproduced by cell division for many generations after the hydrocarbon has been removed.

(5) The population of cells which result show wide morphological variation.

J. C. MOTTRAM.

Mount Vernon Hospital,
Northwood,
Middlesex.
Jan. 1.

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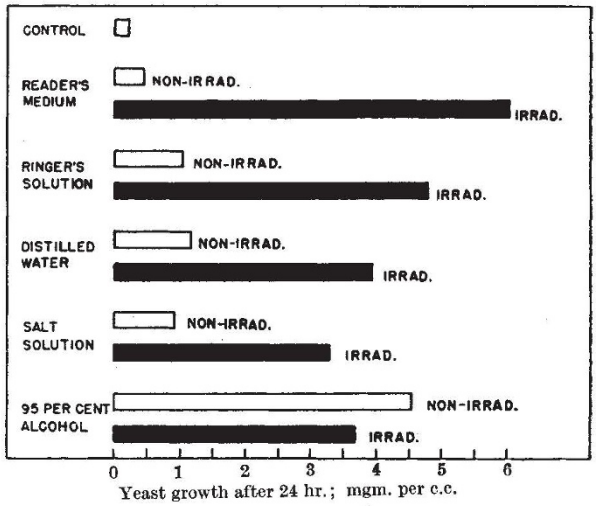
Effect of the Suspending Medium on the Production of Growth Factors by Injured Cells

IN investigating the production of proliferation-promoting intercellular wound hormones by cells injured by various means[1], a study has been made of the effects of the suspending media on the production of the factors by cells injured with lethal ultra-violet radiation

Reasoning that if the proliferation promoters are physiological products of living cells, the ratio of the production of the factors by irradiated and non-irradiated cells should be greatest when they are suspended in a favourable medium, containing nutrient material, a comparison was made of the production of the factors in distilled water, isotonic sodium chloride, Reader's medium[2], 95 per cent ethyl alcohol, and Ringer's glucose phosphate solution. Yeast was suspended in these media at a concentration of 100 gm. per litre. All suspensions were irradiated simultaneously with lethal ultra-violet until there was practically complete killing in the salt solution. The suspensions were then filtered through Berkefeld *N* filters, and the filtrates taken to dryness and made up to five times their original concentrations. These solutions were assayed on yeast according to techniques previously described[3].

The results of typical assays are shown in the accompanying illustration. The greatest production of the factors by irradiated cells and the least by



Yeast growth after 24 hr.; mgm. per c.c.

non-irradiated cells occurred in the most favourable medium (Reader's). Next followed Ringer's glucose phosphate solution. In distilled water and salt solution, in which the cells had no nutrient supply, the ratio of irradiation products to controls, as well as the potency of irradiation products, were less, as would be expected if the factors were elaborated by living cells and not simply extracted from killed cells. There was somewhat greater release of proliferation-promoting factors by both non-irradiated and irradiated cells in distilled water as compared with salt solution, evidently due to the lowered osmotic pressure in the former favouring extraction. In alcohol, irradiation led to less, instead of greater, yields, the toxic effects of alcohol and irradiation together apparently killing the cells too quickly to permit as great elaboration of the factors as in alcohol alone. Were the factors dead-cell disintegration products, one would not expect any decreased yield in alcohol as a result of irradiation unless irradiation destroyed the active factor. That this could not have accounted for the lesser yields obtained was determined by separate experiments on the irradiation of active filtrates, the degree of inactivation being negligible under the conditions of the experiments.

The results indicate that disintegration of dead cells cannot account completely for the proliferation-promoting products obtained from injured cells, and support the hypothesis that injured cells release such factors into their suspending media as a physiological response to injury.

JOHN R. LOOFBOUROW.
Research Laboratories of
Institutum Divi Thomae,
Cincinnati, Ohio.
SISTER CECILIA MARIE DWYER, S.C.
Research Laboratory of
Good Samaritan Hospital,
Dayton, Ohio.
Dec. 7.

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[3] Loofbourow, Dwyer and Morgan, *Studies Inst. Divi Thomae*, **2**, 137 (1938).