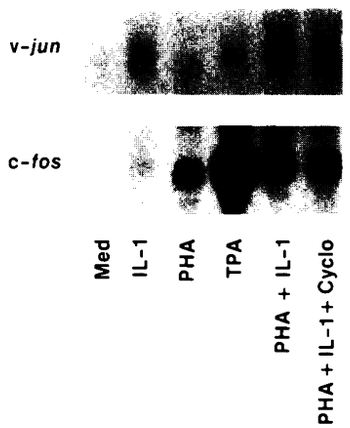

Fig. 2. IL-1 induces c-*jun* whereas PHA induces c-*fos*. LBRM cells were stimulated for 30 min as in Table 1. Cells were incubated in cycloheximide (30 μg/ml) for 30 min (this inhibited 95% of total protien synthesis measured by [^{35}S]methionine incorporation) and stimulated with PHA and IL-1 for another 30 min. Total cytoplasmic RNA was extracted (*27*) and fractionated on a 1% agarose gel (*28*). Blots were hybridized with ^{33}P-labeled mouse c-*fos* or v-*jun* probes. Hybridization with human actin cDNA probes verified uniformity of loaded RNA.

Multiple functional sequences in the IL-2 enhancer have been defined in the past, some specifically responsive to T cell receptor triggering or to TPA stimulation (*6, 11, 18*). We now define a sequence element similar to the AP-1 binding site of the collagenase gene and recognized by a purified AP-1 factor that is responsible for mediating the IL-1 effect on the IL-2 promoter. This element has only weak activity (*7*) in Jurkat cells, a T cell leukemia line that does not require IL-1 to produce IL-2 (it lacks IL-1 receptors). Although the AP-1 site appears to be necessary in LBRM cells, it is probably not sufficient for IL-2 gene expression; TPA alone, like PHA and IL-1, can activate the AP-1 factor but cannot induce the IL-2 gene; this may be because it fails to stimulate the T cell receptor element (−93 to −63) that responds to PHA as a costimulus. In a study of a different cell line, the EL4 thymoma, TPA induced an AP-1 factor that preferentially bound to a different AP-1 binding site on the murine IL-2 promoter (at −150) (*18*). This site, however was not required in LBRM cells, since we deleted the segment from −169 to −101 and retained the IL-1 effect (Table 1A).

TPA and IL-1 have similar actions (*1*) and we show now that IL-1 can act through AP-1 recognition sites. However, there are clearly differences in mode of action between TPA and IL-1. Whereas TPA activates protein kinase C, IL-1 does not (*3*). TPA alone activated the AP-1 trimer, whereas IL-1 required PHA as a costimulus (Table 1C). TPA mainly induced c-*fos*, not c-*jun*, whereas IL-1 induced c-*jun*, not c-*fos* (Fig. 2). TPA can activate the AP-1 factor post-translationally (*19*), and from the current study, it is likely that IL-1 activates AP-1 through de novo synthesis of c-*jun*. Finally, TPA is much less effective than IL-1 as a costimulus for the IL-2 promoter (*4, 8*).

Tumor necrosis factor–α (TNF-α), a cytokine that has actions similar to IL-1, acts via the AP-1 site on the collagenase promoter (*20*). AP-1–like sites occur in many IL-1–inducible genes, including IL-1 itself, IL-6, colony-stimulating factor 1, TNF-α, ornithine decarboxylase, transforming growth factor–β, β nerve growth factor, serum amyloid A, and serum albumin. The AP-1 factor can be produced by most cell types, and thus may provide a general mechanism for the multiple effects of IL-1.

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Human Genotoxicity: Pesticide Applicators and Phosphine

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Fumigant applicators who, 6 weeks to 3 months earlier, were exposed to phosphine, a common grain fumigant, or to phosphine and other pesticides had significantly increased stable chromosome rearrangements, primarily translocations in G-banded lymphocytes. Less stable aberrations including chromatid deletions and gaps were significantly increased only during the application season, but not at this later time point. During fumigant application, measured exposure to phosphine exceeds accepted national standards. Because phosphine is also used as a dopant in the microchip industry and is generated in waste treatment, the possibility of more widespread exposure and long-term health sequelae must be considered.

GLOBAL DEMAND FOR GRAIN AND grain products requires a complex system of transport, storage, and processing before final distribution to the consumer. To maintain the product in a usable state, fumigants and other pesticides are routinely applied by licensed specialists at one or more points from the time of harvest to final processing and use. Before

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1986, carbon tetrachloride and carbon disulfide used in combination accounted for approximately 45% of the fumigant usage in this country (*1*).

Since that time, these well-studied chemical toxicants were formally banned by the U.S. Environmental Protection Agency because of health hazards associated with their use (*2*). The remaining commonly used fumigants are aluminum phosphide, magnesium phosphide, and methyl bromide. Because of ease of application, pellets of aluminum phosphide or magnesium phosphide are most commonly used in the grain industry (*3*). Highly toxic phosphine gas [permissable exposure limit (PEL), 0.4 mg/m^3] is generated from the phosphide; the timed release is dependent on temperature and humidity. Recent work (*4, 5*) demonstrates that phosphine release may take place in as little as 5 min, which places the unprotected worker at risk. Phosphine is also a commonly used dopant in the processing of microchips (*6*). In the hydrosphere, bacterial processing of sewage sludge generates phosphine as a by-product and is a source of environmental contamination (7). Acute lethal effects in insects and humans are associated with the ability of phosphine to inhibit electron transport and combine with heme iron in the presence of oxygen (*8–12*). However, it is only recently that the possible long-term health sequelae of lower level exposure have come under scrutiny. In an epidemiologic evaluation, Alavanja *et al.* reported that grain millers who are exposed to phosphine as well as to other fumigants demonstrated a significant increase in cancer of the hematopoietic system (*3*). Follow-up case control studies suggest that the relative risk of non-Hodgkin lymphoma may be higher than initially reported (*13*).

We began our study by screening human lymphocytes exposed in vitro to the most common fumigants for possible genotoxicity. These in vitro experiments suggested that among the fumigants and pesticides examined, phosphine might be expected to produce a genotoxic effect at low concentrations in vivo. We identified a group of professional fumigant applicators (n = 40) who agreed to participate in our in vivo study. From this group, we selected 24 applicators who met our study criteria. Nine of these applicators were exposed to phosphine alone, 11 to phosphine and other pesticides, and 4 did not use phosphine (Table 1). Community control subjects were selected from a broad category of occupations ranging from banking to skilled labor. These control subjects had no known significant occupational or leisure time exposure to mutagens. Agricultural industry control subjects were chosen from state agricultural department workers who were engaged in the inspection and processing of grain. These workers may be incidentally exposed to pesticides, including phosphine, but are not routinely exposed in their work. Control subjects were matched for age, sex, and to the extent possible, smoking habits. All potential study subjects were interviewed to ascertain health status. The nature and possible consequences of the study were fully explained, and written informed consent was obtained. Only study subjects who fulfilled our study criteria (no chronic disease, no long-term medication use, no recent x-ray) were selected as participants. Diet and leisure activities were also evaluated for significant exposure of the subjects to genotoxic chemicals. Because applicators are generally male, only males were included in the study population.

Immediately before specimen collection, subjects were interviewed again as a check on current health status. Persons with acute or chronic infectious disease were thereby excluded from the study. Because fumigant application is seasonal in the upper Midwest (May through November), specimens were obtained at least twice during the application season, and again 6 weeks to 3 months after the season's work was completed. Where possible, blood specimens were taken from applicators twice within a 24-hour period during times of frequent fumigant application work. One hundred metaphase lymphocytes from each sample were analyzed. We noted no significant intraperson or intersample variation among samples taken and analyzed. Specimens from control subjects were obtained throughout the season within 3 days of receipt of specimens from applicators. This strategy allowed us to compare control samples from different time points with the end of season data collection point. Control subjects from this pool were matched for age and smoking with exposed subjects.

Standard culture and harvest techniques were used. A mixture of RPMI 1640 (Gibco), 20% fetal bovine serum (Hyclone lot no. 1115583), and phytohemagglutin (PHA) (Gibco) (0.75 μg/ml) was routinely used as the culture medium for all cytogenetic studies. The same lot of fetal bovine serum was used throughout the study. Cells were harvested after 48 hours of culture for nonbanded chromosome analysis and stained with 2% Giemsa (*14*). Sister chromatid exchange analysis was performed on

Table 1. Chromosome aberrations were compared among different applicator groups, grain workers, and control subjects. The data from each applicator, consisting of samples studied two or more times during the fumigant application season, were pooled and expressed as the average rate per 100 cells for chromosome aberrations. In each sample, 100 cells were counted per exposed subject. Grain workers and controls were similarly studied over the fumigant application season. The combined data for each subject was converted to a square root scale to homogenize the intersubject variation within groups (*26*). Statistical significance (t test after significant F) was determined by analysis of variance on the converted scale (*27, 28*). In these analyses, exposure-related trends, differences among group means, and possible smoking-related influences were considered. Mean ± SE and range for the untransformed data are presented. Gaps are defined as achromatic regions within a chromatid less than the width of the chromatid. Deletions in the nonbanded preparations are nonstaining regions in a chromatid greater than the width of the chromatid (*29*). Breaks are a discontinuity in a chromatid or chromosome that is misaligned. Rings, dicentrics, quadriradial figures, and acentric fragments result from exchanges within or between chromatids (*30*). Applicators as a group demonstrate significantly increased aberrations compared to grain workers or control subjects with gaps excluded in the computation. Applicators who use phosphine alone compared to applicators who used other pesticides and fumigants but not phosphine and compared to control subjects have significantly increased gaps and deletions. Comparison to the industrial control group (state grain workers) gives similar results.

Mitotic cells counted	Gaps	Deletions	Breaks	Rings, dicentrics, quadriradial figures, acentric fragments	Total (excludes gaps)
		Phosphine alone (n = *9*)			
2400	5.92 ± 1.00 2.5 to 12.3	2.52 ± 0.53 0 to 4.7	1.64 ± 0.28 0 to 3	0.46 ± 0.28 0 to 2	4.62 ± 0.74 1.5 to 8.5
		Phosphine and other pesticides (n = *11*)			
3600	2.86 ± 0.54 0 to 6	1.45 ± 0.48 0 to 4.7	1.67 ± 0.34 0 to 4	0.55 ± 0.16 0 to 1.5	3.67 ± 0.79 1 to 9
		Other pesticides and fumigants (n = *4*)			
800	1.25 ± 0.52 0 to 2.5	1.62 ± 1.01 0 to 4.5	1.25 ± 0.32 0.5 to 2	0.88 ± 0.24 0.5 to 1.5	3.75 ± 0.83 1.5 to 5.5
		State grain workers (n = *15*)			
1500	2.33 ± 0.51 0 to 6	1.20 ± 0.45 0 to 6	0.87 ± 0.32 0 to 4	0.07 ± 0.07 0 to 1	2.14 ± 0.65 0 to 9
		Control (n = *24*)			
2400	3.3 ± 0.51 0 to 8	0.54 ± 0.20 0 to 3	0.71 ± 0.21 0 to 4	0.13 ± 0.09 0 to 2	1.38 ± 0.31 0 to 5

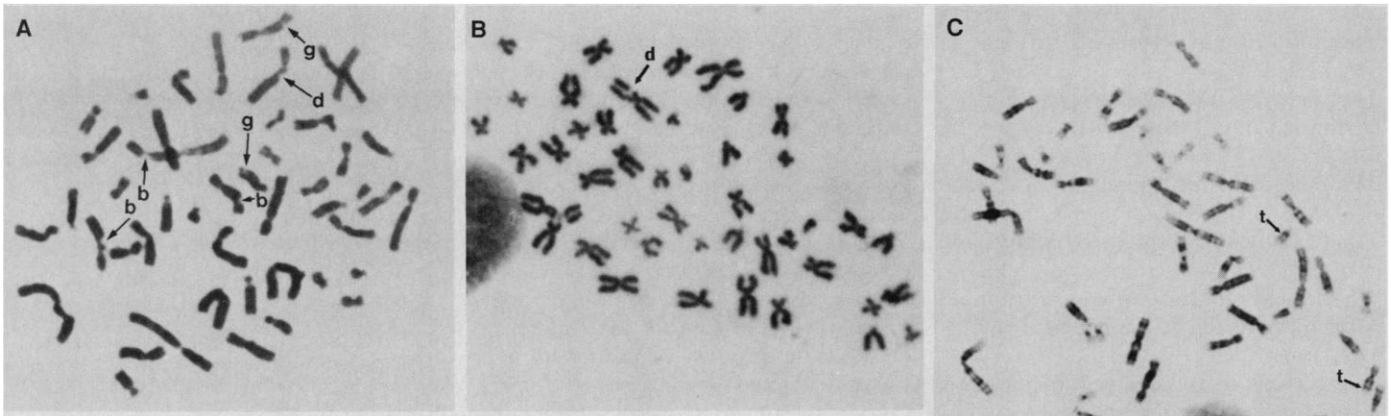

Fig. 1. (**A**) The nonbanded chromosome preparations demonstrate typical chromatid gaps (g), deletions (d), and chromatid breaks (b) observed in phosphine-exposed workers or in lymphocytes treated in vitro with the same chemical (final magnification, ×2200). (**B**) A higher magnification view of chromatid deletion consistently found in applicators who use phosphine is shown. Gaps and deletions are related in vitro to the dose of phosphine added to cultured cells (Table 2) (final magnification, ×2500). (**C**) The Wright-Giemsa banded preparation shows translocation t(10;21)(q24;q22) at the 400-band stage, typical of our preparations. Eleven out of 12 applicators versus 2 of 10 unexposed subjects had chromosome rearrangements, primarily translocations, 6 weeks to 3 months after completion of fumigant work (final magnification, ×2200).

72-hour cultures to which bromodeoxyuridine (BrdU) (8 μg/ml) was added at 24 hours. Cells were stained by the Hoechst + Giemsa method (*15*). All samples were coded and scored.

To index and characterize phosphine exposure, personal breathing zone measurements were made on members of our study group who allowed us access to fumigant application. Among workers involved in enclosed space application (grain bin), exposures ranged from 0.4 to 5.8 mg/m^3 (n = 10) with a mean of 2.97 mg/m^3. Phosphine release from the phosphide occurred in some instances in as little as 5 min. Among workers involved in open air application (rail car), exposure ranged from 0.1 to 0.90 mg/m^3 (n = 4). Phosphine analysis was performed according to National Institute for Occupational Safety and Health (NIOSH) method S-332 (*16*). These data suggested that phosphine exposure during enclosed space application can exceed accepted national PEL standards. These results are similar to those reported by Zaebst *et al.* (*4*).

As a group (Table 1), applicators show 3.58 times more total chromosome aberrations (excluding gaps) than control subjects ($P < 0.001$). State grain workers who may be incidentally exposed to fumigants and pesticides are not significantly different from control subjects in all aberration categories. Phosphine-exposed workers demonstrate an approximately fivefold increase in deletions compared to control subjects ($P < 0.001$). Gaps and breaks are significantly increased among phosphine-exposed workers compared to control subjects ($P < 0.02$ and $P < 0.01$, respectively). Moreover, gaps are significantly higher in the phosphine-exposed group than in any other group evaluated ($P < 0.05$). In workers who used phosphine and other pesticides, or who used other pesticides and fumigants, there were more deletions and breaks than in control subjects ($P < 0.05$). Intergroup comparisons among applicator groups did not show significant differences in breaks or deletions. Phosphine-exposed workers have more than twice as many gaps and deletions than state grain workers ($P < 0.05$) who may be incidentally exposed to phosphine and other pesticides. In the phosphine-exposed groups, most of the gaps and deletions observed involve one chromatid (Fig. 1, A and B). These in vivo data (Table 1) show the same pattern of chromosome aberrations as the in vitro studies (Table 2) that demonstrated a dose-related increase in gaps and deletions due to treatment of lymphocytes with phosphine. The dose range used in these in vitro experiments in culture media is extremely low (Table 2) and is consistent with dose levels that might be expected during exposure to the agent in the application of the fumigant (1 part per million of phosphine = 1.39 μg/liter = 1.38 mg/m^3) (*17*). Cells were harvested later than is usually the case because these in vitro phosphine-induced genotoxic effects are accompanied by mitotic delay. Demonstration in vitro of the same pattern of chromosome aberrations as occurred in vivo further strengthens the association between significant phosphine exposure and increased gaps and deletions. Other agents to which these workers could have been exposed (malathion, methyl bromide, chloropicrin, or carbon disulfide) do not produce this pattern of chromosome aberrations in lymphocytes treated in vitro with these pesticides (*18*). Additionally, partitioning the data on the basis of routine duration of phosphine application (self-re-

Table 2. Chromosome aberration frequencies induced by different doses of phosphine added in vitro. G_0-stage human lymphocytes in complete medium were exposed to phosphine for 20 min at room temperature in gas-tight vials (Pierce Chemical Rockford, Illinois), aerated for one-half hour, and cultured for 96 hours with added PHA. To determine the dose of phosphine in culture medium, tandem cultures were analyzed by gas chromatography for phosphine concentration within one-half hour of addition to culture. The coefficient of variation of replicate phosphine determinations is 3.2%. The nonbanded chromosome aberration frequencies for each phosphine dose are the data from five separate studies. At least 125 mitotic cells per dose were counted. The aberration rate per 100 cells is shown. There is a significant ($P < 0.01$) dose-related increase for gaps, deletions, and total aberrations excluding gaps [linear regression analysis (*31*)]. Details of gas chromatography preparation and analysis are in (*32*). ND, not detectable by gas chromatography.

Dose (μg/liter)	Mitotic cells counted	Gaps	Deletions	Breaks	Rings, dicentrics, acentric fragments	Total (excludes gaps)
Control	200	3.5	0.05	0.1	0	0.15
ND	125	4.0	3.0	2.0	0	5.0
0.26	125	6.4	6.4	0.8	2.0	9.2
1.40	125	7.2	5.6	7.2	1.6	14.4
4.50	125	8.8	10.4	4.0	1.6	16.0

ported) suggests that workers who spend more than 20 min performing this task in an enclosed space ($n = 9$) have more than three times the frequency of gaps and approximately three times more deletions than workers who routinely spend less than 15 min ($n = 7$) performing this task ($P < 0.05$). These data suggest that the absorbed dose of phosphine is proportional to time. Finally, worker protection is highly variable, and exposure without appropriate respiratory protection was common among applicator groups.

In contrast to the findings dealing with chromosome aberrations, there were no significant differences in sister chromatid exchanges between the applicator groups and control subjects. Similar results were obtained in lymphocytes treated with phosphine in vitro.

After completion of the fumigant application season, we attempted to determine whether these chromosome aberrations were persistent in exposed workers. We again obtained specimens from the same group of workers who agreed to continue in the study 6 weeks to 3 months after the end of the fumigant application season. During this time, samples from 12 exposed applicators (6 from the phosphine group and 6 from the phosphine and other pesticides group) were compared to those obtained from 10 nonexposed subjects during the same period. Nonbanded 48-hour cell culture preparations were examined. This methodology captures predominately first division cells containing both stable and unstable aberrations at the induced frequency. Parallel 72-hour nonsynchronized cell culture preparations were banded by a Wright-Giemsa banding technique (*19*). These culture conditions capture second division cells and consequently increase the proportion of aberrations that are stable in vivo and in vitro.

The results from the nonbanded 48-hour cultures showed that the frequency of gaps and breaks in the cultures from workers exposed weeks to months earlier is no different from concurrent controls or from control subjects studied during the application season. These data suggest that the gaps, deletions, and breaks we observed are a transient chromosome abnormality. Moreover, because these postseason nonbanded chromosome data were not different from control subjects, the effect we observed during the fumigant application season (that is, increased gaps and deletions) is not due to seasonal variation in chromosome aberrations (*20*), but is related to phosphine exposure.

Banded analysis of 72-hour cultures demonstrated that 11 out of 12 applicators who used phosphine, or phosphine and other pesticides, had stable chromosome rearrangements in one or more of 100 cells per subject analyzed. Only 2 of the 10 control subjects had such rearrangements. Of a total of 1,200 cells analyzed from the exposed group, 16 (1.3%) showed a stable chromosome rearrangement, including 13 (1.1%) translocations (Fig. 1) and 3 inversions, versus 2 out of 1,000 cells (0.20%) from control subjects. Chromosome rearrangements were more than six times more frequent in the exposed population than in control subjects (χ^2 analysis, $P < 0.05$). Comparison of the frequency of translocation in other banded studies of "normal" human subjects demonstrates some interlaboratory variation. Welch and Lee (*21*) reported a frequency of 0.18% (9 of 5,000); Beatty-DeSana *et al.* (*22*), 0.20% (42 of 21,300); Manolov *et al.* (*23*), 0 of 1,125; and Tawn (*24*), 0.47% (12 of 2,550). Tawn (*24*) indicates that smoking may enhance translocation frequency, but these data include lymphocytes treated with BrdU and analysis of 48-hour cultures. The most common translocation reported involves exchange between chromosome 7 and 14. Our control frequency of 2 out of 1,000 (0.20%) differs significantly (χ^2 with Bonferroni correction) from Beatty-DeSana *et al.* (*22*) [5 of 21,300 (0.02%)] but not from Welch and Lee (*21*) [3 of 5,000 (0.06%)] or Hecht *et al.* (*25*) [4 of 6,000 (0.07%)]. Taken together, the banding data show that applicators who were earlier exposed to phosphine or to phosphine and other pesticides have increased chromosome rearrangements. Because these studies were conducted in 72-hour cultures, it is possible that these rearrangements could have arisen in vitro. However, this is unlikely since cultures from control and exposed subjects were harvested at the same time (72 hours).

Whether the chromosome rearrangements we observed are a specific effect of phosphine is uncertain at the moment. Preliminary analysis and comparison of chromosome breakpoints and rearrangements in banded chromosomes from exposed versus control subjects and of lymphocytes treated with phosphine in vitro suggest this possibility. Larger, more comprehensive studies will be needed to establish whether or not these chromosome rearrangements have long-term biologic significance. Association of breakpoints with oncogene sites, cancer, and specifically, non-Hodgkins lymphoma are possibilities that must be considered.

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31. B. Brown and M. Hollander, Eds., *Statistics: A Biomedical Introduction* (Wiley, New York, 1977), pp. 262–271.
32. Phosphine was generated from magnesium phosphide (Pfaltz and Bauer, Waterbury, CT) dissolved in 78% ethanol in gas-tight vials. Portions (100 μl) of phosphine dissolved in solvent were withdrawn through a Teflon-faced septum and added to cell cultures in a similarly constructed vial. Within one-half hour of addition to culture, phosphine was extracted from the culture and processed for gas chromatography. The gas chromatographic method used is a modification of A. Vinsjansen and K. E.

Thrane [*Analyst* **103**, 1195 (1978)]. In brief, phosphine was extracted from tissue culture medium containing cells with toluene and injected with a gastight syringe into a Varian 3700 gas chromatograph equipped with a nitrogen and phosphorus detector and a Poropak Q column. The detector temperature was set at 250°C. Phosphine standards were prepared from zinc phosphide [D. Hill, in *Analytic Methods for Pesticides and Growth Regulators*, G. Zweig and J. Sherma, Eds. (Academic Press, New York, 1986), p. 145]. The extracted phosphine concentration was determined by comparison to prepared standard concentration curves.

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Electrophysiologic Responses in Hamster Superior Colliculus Evoked by Regenerating Retinal Axons

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Autologous peripheral nerve grafts were used to permit and direct the regrowth of retinal ganglion cell axons from the eye to the ipsilateral superior colliculus of adult hamsters in which the optic nerves had been transected within the orbit. Extracellular recordings in the superior colliculus 15 to 18 weeks after graft insertion revealed excitatory and inhibitory postsynaptic responses to visual stimulation. The finding of light-induced responses in neurons in the superficial layers of the superior colliculus close to the graft indicates that axons regenerating from axotomized retinal ganglion cells can establish electrophysiologically functional synapses with neurons in the superior colliculus of these adult mammals.

THE FAILURE OF CUT AXONS TO REgenerate in the central nervous system (CNS) of adult mammals causes a permanent functional disconnection between the axotomized neurons and their targets. However, when transplanted segments of peripheral nerve (PN) are used in vivo to test and influence the regenerative capacities of axotomized CNS neurons, nerve cells in various regions of the CNS can regrow lengthy axons (*1*). When the optic nerve of adult rodents is replaced with a PN graft joining the eye and the superior colliculus (SC), the regenerating axons of some axotomized retinal ganglion cells (RGCs) extend for several centimeters through the grafts, penetrate the SC for short distances, and form well-differentiated synapses in the superficial layers of the SC (*2, 3*). At least some of the RGCs that regrow axons into PN grafts can respond to visual stimuli and must therefore have afferent connections from other neurons within the retina (*4*). There has been no proof, however, that the regenerating axons of these or any other adult mammalian CNS neurons can transsynaptically excite or inhibit neurons in the reinnervated area of the CNS.

To investigate the possibility that contacts between regenerated RGC axons and SC neurons could mediate synaptic transmission, we recorded unitary responses to light in the SC of adult hamsters with cut optic nerves and PN grafts linking one retina and the ipsilateral SC (*5*). Transection of both optic nerves ensured that any responses to light recorded in the SC would be mediated by RGC axons that had regenerated across the PN graft. In addition, we took care to distinguish postsynaptic responses of SC neurons from responses in RGC axons that had penetrated the SC. These electrophysiologic experiments were performed 15 to 18 weeks after the grafts were implanted in the SC.

Extracellular recordings with glass-insulated carbon fiber electrodes (*6*) were made in the right SC of eight animals with PN grafts while a high-intensity, 10-μs flash of light was delivered every 3.1 s by a Grass photic stimulator 8 to 10 cm from the eye with the graft (*7*). For purposes of comparison, responses to the flash of light were recorded directly from regenerated axons of RGCs in one hamster. The caudal end of the graft was removed from the SC 14 weeks after insertion, and small bundles of axons were teased from the graft and placed on a silver wire electrode with a nearby ground as a reference (*4*).

Excitatory or inhibitory responses to the flash of light were recorded from single units in the SC of all eight animals. Responsive units tended to be clustered together. When a unitary response to the flash was obtained (Fig. 1A), the following procedure was used in an attempt to establish that the response was recorded from an SC neuron rather than from a regenerated RGC axon that had grown into the SC. Units that responded to light were identified as transsynaptically activated SC neurons if they also responded to electrical stimulation of the graft at a variable latency with an ill-defined threshold (Fig. $1B_1$) and if stimulation of the graft with double (or in some cases triple) pulses separated by 2 to 4 ms evoked one or more spikes more consistently than single pulses at the same intensity (Fig. $1B_2$). Sequential subthreshold postsynaptic potentials of appropriate timing

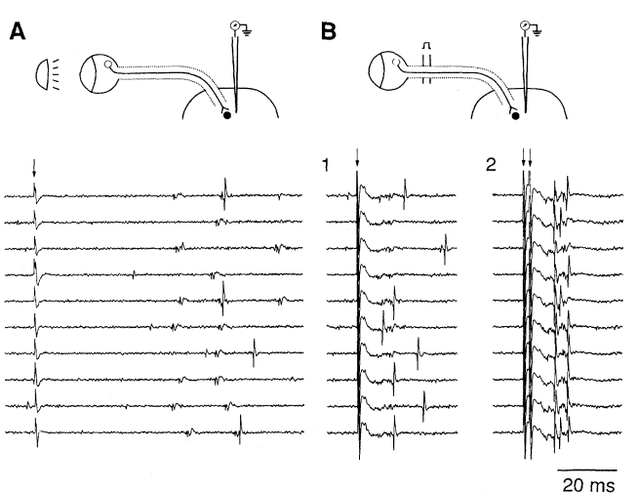

Fig. 1. (**A**) Ten successive responses to light flash (at arrow) recorded at a depth of 250 μm in the SC. The large unit responds with a single spike on four of ten iterations. (**B**) The same unit responds erratically with inconstant latency to (traces 1) single electrical stimuli (arrow) delivered to the graft and responds at a more constant latency, often with multiple spikes, to (traces 2) paired electrical stimuli (arrows) of the same intensity. This pattern of response is inconsistent with that anticipated from an RGC axon and thus identifies this unit as an SC neuron. All recordings were filtered (band pass 500 Hz to 5 kHz) and stored on magnetic tape.

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