EFFECTS OF MARIJUANA
ON HUMAN NATURAL KILLER CELL ACTIVITY

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INTRODUCTION

Marijuana and its major psychoactive component delta-9-tetrahydrocannabinol (THC) were first reported to depress various immune functions in humans as early as the mid 1970's. (Reviewed in 1). However, these studies yielded conflicting data necessitating further investigations. Recent studies by Friedman and co-workers confirmed the immune suppressive effects of THC in mice using in vivo and in vitro experimental models (2-5). Klein et al. (6) reported depression of murine natural killer (NK) cell function in mice following in vitro exposure to THC while Patel et al. (7) demonstrated inhibition of rat NK cell cytotoxicity. A re-examination of responses of human peripheral blood mononuclear cells (PBMC) following exposure to THC in vitro indicates that lymphocyte blastogenesis (8), monocyte/macrophage spreading on plastic surfaces and phagocytosis (1) and NK cell activity (9,10) all are diminished in the presence of the drug.

The focus of this review of our studies will be upon the conditions under which THC depresses NK cell function and some of the mechanisms involved in this effect.

METHODS AND RESULTS

Conditions for Inhibition of Natural Killer Cell Activity

Human PBMC were collected by venipuncture, often obtained from a unit of blood. The buffy coat was separated from whole blood and then mononuclear cells were isolated using Ficoll-Hypaque. Initially PBMC labelled with Na$^{51}$CrO$_4$ were used to determine cytotoxic concentrations of THC for the leukocytes. THC was shown to be toxic, causing the release of chromium at 20 $\mu$g/ml but not at lower concentrations when cells were exposed to drug for 3 hr (9). Subsequent studies indicated that cell viability was unaffected when mononuclear cells were exposed to 10 $\mu$g THC/ml for 48 hr (Specter et al., unpublished observations).

NK assays were performed as described in Specter et al. (9). PBMC were incubated with K562 target cells previously labelled with $^{51}$Cr. After a 4 hr
Cultures of lymphocytes were exposed to THC for 0 to 18 hr prior to their use in the NK assay. These cells were then incubated with the target cells in the standard NK assay. Experimental controls consisted of cell cultures incubated with medium alone or medium containing 0.1% dimethyl sulfoxide (DMSO), the diluent for THC.

The toxic THC concentration of 20 µg/ml, when incubated with effector cells for 1 hr or longer before initiation of the NK assay, virtually ablated all NK activity. However, little effect was noted when THC was added at the beginning of the NK assay (time 0) (Figure 1). THC at 10 µg/ml was not toxic to the cells, and suppressed NK cell cytotoxicity after a 1 hr incubation with drug before initiation of the NK assay. The inhibitory effect was temporally related, with increasing suppression of NK activity observed as time of pre-incubation with THC was lengthened. Lower concentrations of THC had a diminished effect (data not shown) and 1 µg THC/ml had no effect on NK cell killing even after a 4 hr exposure of the lymphocytes to the drug. In parallel experiments cells were exposed to THC for 3 hr and then washed to remove unabsorbed THC before testing in the NK assay. This procedure did not reduce the THC effects described in Figure 1 (Table 1). Subsequent experiments incorporated washing effector cells to ensure that none of the THC effects were on target cells. In all subsequent experiments PBMC were incubated with THC for 3 hr as standard prior to testing in the NK assay, unless specified otherwise.