Prostaglandin E2 administered via anus causes immunosuppression in male but not female rats: A possible pathogenesis of acquired immune deficiency syndrome in homosexual males

(Semen/cellular immunity/sex-related difference)

Sachiko Kuno, Ryuji Ueno, and Osamu Hayashi

Hayashi Bioinformation Transfer Project, Research Development Corporation of Japan, c/o Osaka Medical College, 2-7 Daigakumachi, Takatsuki, Osaka 569, Japan

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ABSTRACT To explain a possible pathogenesis of acquired immune deficiency syndrome (AIDS) in homosexual males, we propose the following hypothesis. Prostaglandin E2 in human semen given via anus during anal intercourse may cause an immune dysregulation in the male semen recipients; this immunosuppressive effect of prostaglandin E2 may be one of the underlying factors that stimulate AIDS-associated virus infection or that trigger the latent AIDS-associated virus. This hypothesis is supported by the following experimental results. Anal infusion of prostaglandin E2 into male rats reduced in vitro responses of T lymphocytes to phytohemagglutinin. However, the T-cell response of female rats was not reduced significantly by the anal infusion of seminal prostaglandins.

Although versatile actions of prostaglandins (PGs) in various organs have been found, PGs were originally discovered in human semen in the 1930s (1, 2). The concentrations of seminal PGs are extraordinarily high (3, 4), and physiological roles of PGs in semen have been investigated mainly with respect to reproduction (5, 6). The recent emergence of acquired immune deficiency syndrome (AIDS) led us again to pay attention to PGs in human semen. AIDS is thought to be caused by a pathogenic virus referred to as human T-lymphotropic virus type III (HTLV-III) (7), lymphadenopathy-associated virus (LAB) (8), and AIDS-associated retrovirus (ARV) (9). However, accumulation of epidemiological information revealed a strict predilection for the severe disease in people with selected risk factors (10, 11). Especially in homosexual males, the highest risk group for AIDS, immunological abnormalities were often observed without clinical signs of the disease (12–15) and, in many cases, without serum HTLV-III antibody (16). These findings suggested that the infectivity or activity of AIDS-associated virus might be dependent on the immunity of the host. PGF2 and PGD2 are known to suppress immune functions in vitro (17, 18). We speculated that seminal PGs given to receptive homosexual males might induce immunosuppression. These immunosuppressed people may be more susceptible to the viral infection leading to AIDS. To examine the above hypothesis, we investigated effects of PGs administered by anal infusion on cellular immunity in both male and female rats.

MATERIALS AND METHODS

Chemicals. PGE2, -D2, and -F2a were kindly supplied by Ono Central Research Institute (Osaka, Japan). Other chemicals were purchased from Nakarai Chemicals (Kyoto, Japan) and were of reagent grade.

Animals. Specific-pathogen-free male and female rats of the Wistar strain (7–8 weeks old) were purchased from Kitayama Labes (Kyoto, Japan) and kept in a soundproof room at 24°C and 50 ± 5% relative humidity on a 12-hr light/dark cycle.

Administration of PGs. A stock solution of PGE2, -D2, or -F2a (50 mg/ml in ethanol and stored at −20°C) was diluted 1:200 with phosphate-buffered saline (137 mM NaCl/2.68 mM KCl/8.1 mM Na2HPO4/1.47 mM KH2PO4, pH 7.4) containing fructose at 5 mg/ml. After incubation at 37°C for 5 min, this solution was infused at 5 cm from the anus of male or female rats each day (between 1000 and 1200) for 7 days. The same volume of buffer containing 0.5% ethanol was administered to control rats.

Determination of Phytohemagglutinin (PHA) Response of Peripheral Blood Mononuclear Cells. Three hours after the final infusion of PG, rats were anesthetized by intraperitoneal injection of pentobarbital (30 mg/kg of body weight), and 7 ml of blood was collected from the right ventricle of the heart. Blood samples were immediately transferred into sterile tubes containing 2 ml of heparinized Hanks balanced salt solution and sent to Special Reference Laboratories (Tokyo, Japan) for the analysis of blastogenesis of T lymphocytes against PHA. The following experiments were performed in Special Reference Laboratories according to the method of Bøyum (19). These experiments were done within 10 hr after sampling of blood. Peripheral blood mononuclear cells were separated from blood samples, and the final cell concentration was adjusted to 5 × 10⁵ cells per ml. This preparation was cultured with or without PHA (30 μg/ml) at 37°C for 64 hr under 5% CO2 in a culture medium with 12 mM Hepes buffer (pH 7.2), penicillin (100 units/ml), streptomycin (100 μg/ml), and 10% fetal bovine sera. Eight hours before harvesting, 0.25 μCi (1 Ci = 37 GBq) of [3H]thymidine was added to each well (240 μl per well) of a 96-well plate. These cultures were harvested on glass filter papers for the measurement of [3H]thymidine incorporation into DNA by liquid scintillation. Each value of [3H]thymidine incorporation represents the mean of triplicate cultures from one sample.

Determination of Sensimal PGs. Human semen samples were collected from seven healthy volunteers [age 32.3 ± 4.2 years (mean ± SD)]. Extraction and radiioimmunoassay of PGs were carried out by the method of Narumiya et al. (20). The crossreactivities of PG-specific antisera to other PGs were <1%.

RESULTS

PGE2, -D2, or -F2a (0.5 mg/kg) was administered via anus to male or female rats once a day for 7 days, and in vitro

Abbreviations: AIDS, acquired immune deficiency syndrome; PG, prostaglandin; HTLV-III, human T-lymphotropic virus type III; PHA, phytohemagglutinin.
blastogenic responses of peripheral blood mononuclear cells to PHA, a mitogen for T-lymphocytes, were investigated by measuring [3H]thymidine incorporation (Fig. 1). In male rats, the blastogenic responses to PHA were markedly reduced by the anal administration of PGE2 or -D2, whereas PGF2α did not show any significant effect on the mitogen response. Similar reduction of blastogenic responses to PHA was observed after anal administration of PGE2 at 0.25 mg/kg or of PGD2 at 0.25 or 0.05 mg/kg, but PGE2 at 0.05 mg/kg was almost ineffective. In contrast to males, female rats infused with PGE2 at 0.5 mg/kg showed a slight increase in the blastogenic response to PHA, and the immunosuppressive effect of PGD2 was less significant. PGF2α had no effect on PHA response in female rats.

The concentrations of PGE2 and PGF2α in human semen have been determined (3, 4), but no studies on the amount of PGD2 have been reported. We determined the concentration of PGD2 in addition to PGE2 and -F2α in human semen by radioimmunoassay. Although PGD2 had a potent immunosuppressive effect on male rats, human semen was almost devoid of PGD2 (<0.2 μg/ml). On the other hand, PGE2 was the major PG among the three PGs in human semen, a result was confirmed by gas chromatography/mass spectrometry (unpublished data). The concentration of PGE2 in human semen was 61 ± 15 μg/ml (mean ± SEM, n = 7) in this study. The concentration of PGF2α was 5 ± 1 μg/ml.

**DISCUSSION**

There is much evidence, from in vivo and in vitro experiments, that PGE plays a role in immunoregulation (17, 21). Most of this evidence supports the idea that PGE suppresses both cellular and humoral immunities. Further, it has been reported that PGE2 induces or activates suppressor T cells, which release suppressor factors to inhibit the response of T lymphocytes to mitogens (22, 23). A physiological role of the immunosuppressive effect of PGE2 in vivo is thought to be necessary to prevent an overreaction of the immune system (17). Therefore, if exogenous PGE were administered repeatedly at high concentrations, it would cause an unnecessary immunosuppression in the recipients.

Our results show that PGE2 infused via anus caused a decrease in T-cell response to PHA in male rats. Genital-anal intercourse is a common sexual activity among homosexual males, indicating that seminal constituents are absorbed by the receptive partners, one of the highest risk groups for AIDS (10). It is not clear whether the underlying immunosuppression is essential for the infection and/or development of AIDS-associated virus. However, the immunosuppression in homosexual males that is reported even in apparently healthy (12–15) or HTLV-III-antibody-negative groups (16, 24) may arise from seminal PGE2 received during anal intercourse. In theory, these immunosuppressed males might be more susceptible to the viral infection, although the possibility is not excluded that other factors, involving repeated immunization against sperms as alloantigens (25, 26), additively cause the immunosuppression in homosexual males. A sex-related difference of the sensitivities to PGE2 and -D2 was observed. One possible explanation is that females should be naturally more resistant to seminal immunosuppressants such as PGE2. It seems reasonable that females have special protection against seminal PGs. Further, the immunity of females is originally superior to that of males because of X-linked genetic (27) and hormonal (28) regulation. The difference in sensitivities to immunosuppressive PGs between males and females might explain why male semen recipients are included in the highest risk group for AIDS.

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