Discussion. Sawanobori and co-workers[8] investigated the subpopulations of peripheral blood T and B lymphocytes in 11 patients with recurrent herpes genitalis utilizing SRBC rosette tests and described that patients with herpes genitalis had normal percentage of early rosettes (active T lymphocytes), total rosettes (total T lymphocytes) and EAC rosettes (total B lymphocytes). Our findings presented herein indicate, in contrast to those by Sawanobori et al., that percentages of active T lymphocytes are apparently lower than those of normal controls. This differene is, at least in part, due to the analytical methods employed; the previous authors used a SRBC: lymphocytes ratio of 8:1, while we used that of 27:1. Furthermore, the patients they studied were subject to recurrent herpes genitalis, whereas 15 of 16 patients in our series had no episode of herpes genitalis or labialis despite the presence of serum neutralizing antibodies[9,10]. Approximately 95% of Japanese females aged 31 or over carry serum neutralizing antibodies against either HSV-1 or HSV-2 without apparent episode of herpes labialis or genitalis. Accordingly, clinical or immunological features of herpes genitalis in Japanese women seem to be different from those in Caucasian or Negro women. The viral types involved in herpes genitalis also differ between Japanese women and Caucasion or Negro women. More than two thirds of the occurences of herpes genitalis in the USA are due to HSV-2, while in Japan approximately half are due to HSV-1[11,12]. These methodological or epidemiological differences may be responsible for the different results in the 2 studies, and our present findings agree with the description by Wybran and Fudenberg that all patients with viral disease had less than 15% rosette forming cells in the active T cell assay[13].

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Dietary immunostimulation: Interaction with BCG and LPS

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Summary. The action of BCG and LPS mR595 used in conjunction with a formula-defined diet is dependent on the administration timing and resembles that of interacting adjuvants affecting different elements of the immune system.

Production of anti-sheep erythrocyte (SRBC) antibody has been shown to be enhanced in rats fed an easily absorbed formula-defined diet (FDD), instead of the usual laboratory chow[2]. Such formulations may be used, in the clinic, to lessen the nutritional imbalance resulting from cancer and cancer therapy[3]. On the other hand, over the past years, BCG has been used as an adjunct against cancer in humans[4-6], and anti-tumour properties of bacterial lipopolysaccharides (LPS) have been investigated[7,8]. However, the fact that FDD may influence the immune response suggests that the action of adjuvants administered to FDD-fed organisms could be analogous to the interaction of different immunostimulants. Present studies were initiated to verify this hypothesis.

Virgin female Sprague-Dawley rats, 6–7 weeks old, were fed FDD 3[2] or Purina laboratory chow 5001 for 7 days, and challenged with SRBC alone or in conjunction with BCG (bovine strain, live, 1 mg i.p.) or LPS mR595 (*Salmonella minnesota,* 40 µg i.p.). Both concentrations were used in view of their known oncotherapeutic action[4,8]. Since the administration timing is known significantly to affect their action[4,7], the adjuvants were given either together with (concomittantly) or within minutes after (sequentially) SRBC. Logarithmic transformation of the titers was performed and mean titer values were obtained. Differences between the means were appraised using Student's t-test (N-2 degrees of freedom, 2-sided level of significance).

As previously reported, the FDD alone was responsible for more than a 7-fold increase of the anti-SRBC antibody titers (table). Administered sequentially, BCG provided no further immunostimulation than that provided by the diet ($p > 0.1$) while a 25-times increase was observed in the chow-fed controls ($p < 0.001$). As to sequential LPS mR595, the 4–5-fold enhancement ($p < 0.05$ for each dietary group) achieved was diet-independent, but did not hinder the stimulating action of the FDD. In fact, the LPS and the FDD had a compounded action. Administered concomittantly, the LPS was totally ineffective in both dietary groups ($p > 0.1$), while BCG provided a significant ($p < 0.01$ for both groups) and food-dependent antibody increase (6- and 3-fold, respectively in chow- and FDD-fed animals).

The adjuvant action of BCG and LPS mR595 is influenced by both the administration timing and the types of food eaten. Known to stimulate the reticuloendothelial system and the T-cells[9], BCG can also act similarly to LPS[9,10] which substitute for T-cells and stimulate B-lymphocytes[11,12]. This enables BCG to affect both cellular and humoral immunities while lipopolysaccharides influence primarily the humoral response. As to FDD3, it increases

cellular activity in the bone marrow and the lymph nodes[13,14] and enhances humoral immunity[15], suggesting an adjuvant-like behaviour. Previous data account also for the differences observed in the action of BCG or LPS mR595 in the 2 dietary groups.

The influence of the FDD on the immune system is known to be multileveled and sex-related[2]. From the present data, the following conclusions can be drawn: a) Lipopolysaccharides are a diet-independent class of adjuvants, in contrast to BCG; b) FDD3 alone provides only intermediate immunostimulation. When adjuvants are used in conjunction with the diet, proper agent and administration timing are required to achieve full stimulation; c) given the right conditions, the action of BCG may be equivalent to the combined effects of LPS mR595 and FDD3. Our results suggest that using BCG or lipopolysaccharides in conjunction with formula-defined diets may be analogous to dealing with interacting adjuvants affecting different elements of the immune system.

Action of formula-defined diet 3 on the immune system response to SRBC in the female rat: Effect of sequential or concomittant BCG and LPS mR595

Treatment	Anti-SRBC[a] antibody levels in serum[b]	
	Animals fed laboratory chow	Animals fed diet 3
SRBC	3.5 ± 0.5[c] 11[d] (8-16)[e]	6.5 ± 0.4 90 (69-119)
SRBC + BCG^{f}_{seq}	8.2 ± 1.2 294 (128-657)	6.5 ± 0.2 91 (78-105)
SRBC + LPS $mR595^{g}_{seq}$	5.9 ± 0.5 59 (43-80)	8.4 ± 0.3 337 (274-416)
SRBC + BCG_{con}	6.2 ± 0.6 72 (48-109)	8.2 ± 0.5 288 (207-402)
SRBC + LPS $mR595_{con}$	4.5 ± 1.0 23 (12-44)	6.5 ± 0.56 91 (61-133)

[a]Sheep erythrocytes (1 ml, 10 cells, injected i.p. [b]Determined by passive hemagglutination, 7 days after SRBC challenge. [c]Mean number of wells (x) ± SE (6 animals per group, repeated once). [d]Titer as obtained by 2^x. [e]Titer range as obtained from SE of x. [f]Bacillus Calmette-Guérin (Institut Armand Frappier, Laval des Rapides, Québec, Canada) 1 mg/animal, injected i.p. [g]Lipopolysaccharides of *Salmonella minnesota* mR595 (provided by Dr O. Lüderitz, Max-Planck-Institut für Immunobiologie, Freiburg, FRG), 40 µg/animal, injected i.p.

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Release of gastrointestinal hormones following an oral water load

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Summary. The ingestion of 2 different water loads (7.5 and 15 ml/kg) by healthy subjects stimulated the release of plasma motilin, gastrin, pancreatic polypeptide and VIP. Atropine was found to block the release of PP but not the other hormones.

The mechanisms involved in the postprandial release of intestinal hormones are ill understood. The nutriment component of food is clearly important but the role of other stimuli such as water ingestion have not been previously investigated. This study provides evidence that it results in the release of several intestinal hormones.

Subjects and methods. All subjects were healthy and were studied after an overnight fast in 2 groups. a) 7.5 ml of distilled water (temperature 20 °C)/kg b.wt was given orally to 6 healthy volunteers (aged 25-35 years, weighing 70-85 kg). b) 15 ml water/kg b.wt was given to a 2nd group of 6 healthy subjects (aged 26-34 years, weighing 75-85 kg). c) The last experiment was repeated 2 weeks later on 5 of the group (b) subjects who received 600 µg atropine i.v. 2 min prior to ingestion of the water load (15 ml/kg). The mean ingestion time of the water for both groups was 2 min.

Hormone radioimmunoassays were performed with conventional methodology using antisera raised to human gastrin[2] and pancreatic polypeptide (pp)[3] to porcine gastric inhibitory polypeptide (GIP)[4], vasoactive intestinal polypeptide (VIP)[5], glucagon[6] and motilin[7] and to bovine insulin[8]. The assays could detect the following changes in plasma hormone concentration with 95% confidence: gastrin 2 pmoles/l, PP 2 pmoles/l, GIP 8 pmoles/l, enteroglucagon 10 pmoles/l, pancreatic glucagon 2 pmoles/l, VIP