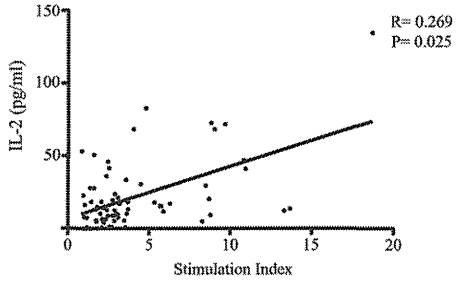

FIG E2. IL-2 concentrations in culture supernatant of cow's milk protein–stimulated PBMCs correlated significantly with antigen-specific lymphoproliferation. PBMCs from children with gastrointestinal food allergies were stimulated separately with 100 μg/mL of each of 5 LPS-depleted milk protein preparations in the absence of serum for the antigen-specific lymphoproliferation assay and in the presence of 5% autologous plasma for the IL-2 production assay. The stimulation index was calculated as milk protein–specific tritiated thymidine uptake (cpm)/vehicle-induced tritiated thymidine uptake (cpm), and the highest stimulation index shown among the 5 tested protein preparations was used as that patient's data in the plot. Even under slightly different culture conditions, antigen-specific lymphoproliferation and antigen-specific IL-2 production were significantly correlated ($r = 0.269$, $P = .025$).

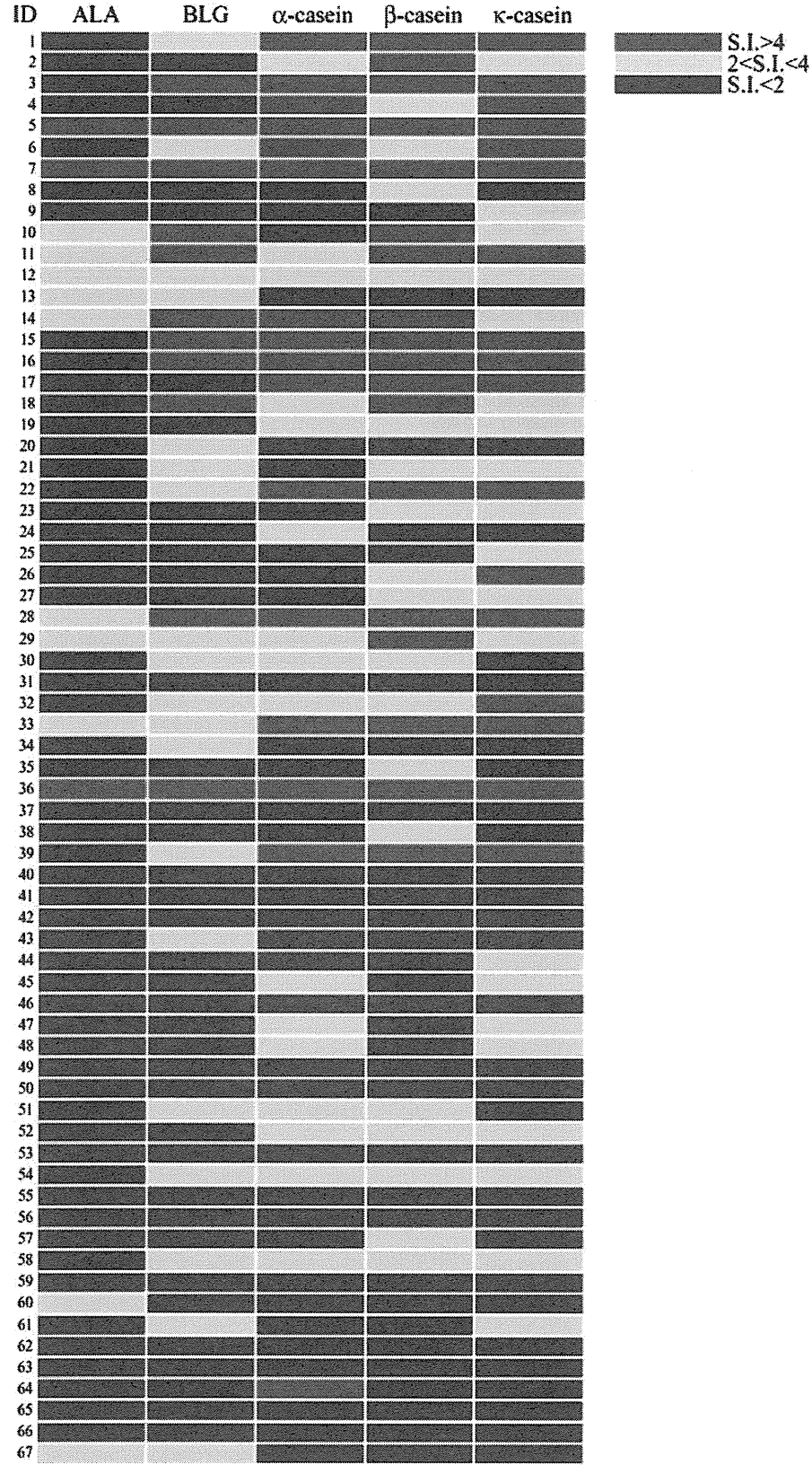

FIG E3. The milk protein component causing the most prominent tritiated thymidine uptake varied among the patients. PBMCs from children with gastrointestinal food allergies were stimulated separately with 100 μg/mL of each of 5 LPS-depleted milk protein preparations in the absence of serum. Lymphoproliferation was measured based on tritiated thymidine uptake. The stimulation index *(S.I.)* was calculated as milk protein–specific tritiated thymidine uptake (cpm)/vehicle-induced tritiated thymidine uptake (cpm). For

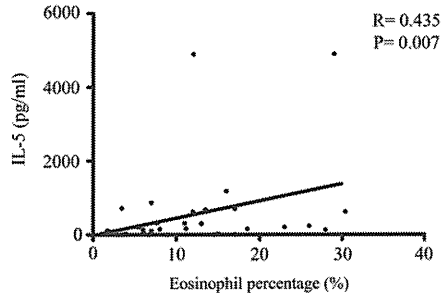

FIG E4. IL-5 concentration in the culture supernatant of cow's milk protein–stimulated PBMCs correlated significantly with the peripheral blood eosinophil percentage. PBMCs from children with gastrointestinal food allergies were stimulated separately with 100 μg/mL of each of 5 LPS-depleted cow's milk protein preparations in the presence of 5% autologous plasma for 6 days. Antigen-specific IL-5 production correlated significantly with the peripheral blood eosinophil percentage at disease onset (r = 0.435, P = .007).

each patient, the SI is shown for the PBMCs' response to each of the 5 milk protein preparations. Each *row* represents a single patient, and each *column* represents one of the 5 milk proteins. *ALA*, α-Lactalbumin; *BLG*, β-lactoglobulin; *blue*, SI < 2; *yellow*, 2.0 < SI < 4.0; *red*, SI > 4.

TABLE E1. Concentrations of LPS in commercially available milk protein preparations before and after treatment with a prepacked endotoxin affinity column

Cow's milk protein preparation	Before treatment (pg/mg)	After treatment (pg/mg)
α-Lactalbumin (Sigma L-6010)	184,200	14
β-Lactoglobulin (Sigma L-3908)	206,700	1,880
α-Casein (Sigma C-6780)	540	23
β-Casein (Sigma C-6905)	500	34
κ-Casein (Sigma C-0406)	400	41
LPS-depleted β-lactoglobulin (Bean Stalk Snow)	29	—

The indicated milk protein preparations were treated with a prepacked endotoxin affinity column (Detoxi-Gel; Pierce Chemical, Rockford, Ill) in accordance with the manufacturer's instructions. LPS concentrations were measured by using the limulus amebocyte lysate assay.