Changes in *Brassica napus* Progoitrin Induced by Sheep Rumen Fluid

Sir: Progoitrin, a thioglucoside contained in the seeds and defatted meal of *Brassica napus*, is known to have toxic effects, through its antithyroid and, therefore, goitrogenic action, upon monogastric animals, while it is virtually harmless for ruminants.

Under the effect of myrosinase,—an enzyme found in *B. napus*, progoitrin is converted into a variety of substances, depending upon pH (M.A. Greer, *Arch. Biochem. Biophys.* 99:369 [1962]; M.E. Daxenbichler, C.H. Van Etten, W.H. Tallent, and I.A. Wolf, *Can. J. Chem.* 45:1972 [1967]; A. Lanzani and G. Jacini, *Riv. Ital. Sostanze Grasse* 50:277 [1973]), the most commonly known of which is 5-vinyl-1,3-oxazolidine-2-thione (VO).

In a previous article (A. Lanzani and G. Jacini, *Riv. Ital.* Sostanze Grasse 48:471 [1971]), it was reported that VO, when treated under such conditions as were described by T. Mukaiyama, I. Kuwajima, and K. Mizui (J. Org. Chem. 31:32 [1966]) for 3-phenyl-1,3-oxazolidine-2-one, undergoes a similar rearrangement, yielding the isomer 5-vinyl-1,3-thiazolidine-2-one (VT) and a polymer:

$$CH_{2}-CH_{0}-CH_{0}-CH_{0}-CH_{2}-CH_{0}-CH_{2}-CH_{0}-$$

The purpose of this letter is to report that the VT also is formed from progoitrin (whether pure or contained in *B. napus* meal) by an enzymatic process when it comes in contact with sheep rumen fluid. In addition to VT and VO, other substances are formed, which currently are being investigated.

We have put sheep rumen fluid in contact at 39.5 C with (A) *Brassica* meal containing 3.4% progoitrin, (B) a mixture of cellulose and progoitrin in the same percentage, and (C) progoitrin-free alfalfa meal. In one series of experiments, the sheep had been kept on a diet containing no *B. napus*, whereas in another series the animal had been accustomed to a diet containing *B. napus* meal.

After 2 and 24 hr, each mixture was lyophilized; then

TABLE I

Transformation Products from Progoitrin by Sheep Rumen Fluid

Percent of lyophilized residue	Rapeseed meal containing substrate		
	Oh	2h	24h
Unreacted progoitrin	0.28	0.02	0
as VO	(0.28)	(0.12)	(0)
Newly formed VO	0	0.02	0.02
	(0)	(0.03)	(0.09)
VT	0-	0.02	0.08
	(0)	(0.02)	(0.06)

All data in parentheses related to results obtained with rumen fluid of sheep that had been fed no rapeseed meal.

L.A. Appelqvist's and E. Josefsson's method (J. Sci. Food Agr. 18:510 [1967]), involving enzymatic conversion of progoitrin to VO and measurement of the latter by UV, was used to determine such residual progoitrin as remained in the dry residue, as well as the amount of VO formed in the process (residual progoitrin being calculated from the difference between total VO and preformed VO found by the analysis). Other transformed products, if any, also were investigated. Comparable results were obtained when rumen juice was put in contact with progoitrin in B. napus meal and with the pure compound mixed with cellulose.

Table I evidences a gradual decrease in progoitrin, until the substances disappeared altogether after 24 hr, being replaced by several transformation products: VO, VT, and other still unidentified substances.

When checks were performed on alfalfa meal under identical conditions, none of the mentioned substances could be isolated.

In a parallel sequence of tests, where rumen juice was put in contact with cellulose to which VO, instead of progoitrin, had been added, the latter was found to undergo a transformation, without, however, yielding any VT (Table II). In this case too, transformation products currently are being investigated. Such substances as were obtained in the course of all the above reactions were separated by thin layer chromatography, using hexane-ethyl ether (1:1) as the eluent and identifying both VO and VT by UV and IR spectrophotometry (A. Lanzani and G. Jacini, Riv. Ital. Sostanze Grasse 48:471 [1971]). Consequently, the assumption would seem justified that progoitrin is transformed by rumen juice into other substances, among which VT prevails. This content is substantiated indirectly by a recent work of A. Rutkowski, H. Kozlowska, and J. Cwik (Riv. Ital. Sostanze Grasse 50:16 [1973]).

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TABLE II

VO Transformation by Sheep Rumen Fluid

Percent of lyophilized residue	Cellulose + 1% VO		
	Oh	2h	24h
vo	0.1 (0.1)	0.01 (0.02)	0 (0.01)
VT	0	0	0

Data in parentheses were obtained with rumen fluid from sheep that had been fed no rapeseed meal.

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