Canola Check Sample Series

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ABSTRACT

Four separate check series for canola seed, canola meal, crude and crude degummed canola oil and refined, bleached and deodorized canola oil were started in July 1980. They are devised to help laboratories calibrate analytical procedures to be used on canola products. This paper describes the organization of the program and special features of canola analysis. It indicates and discusses less than reliable analyses and describes the active participation of the program organizers to improve reliability of analyses.

INTRODUCTION

In 1981, canola oil comprised 50.7% of refined oil processed in Canada and 70.5% of the salad oil. As a comparison, refined soybean oil was 29.3% of the total (1). Even though there is this dominance of canola in Canadian oilseed preference, most Canadian laboratories have not in the past had any check sample series devoted to canola, other than an erucic acid series. Canadian laboratories have relied on internal checks or on the AOCS Smalley series. The Smalley series coordinators have been very accommodating in including rapeseed and canola samples but they have naturally not been able to emphasize an oilseed which is grown little in the United States.

Canola seed differs from soybean or cottonseed in many particulars such as size, color, fatty acid composition, oil content, presence of glucosinolates and absence of trypsin inhibition. Thus, methods appropriate to soy or even flax do not necessarily apply. After consultation with industrial and government laboratories, we applied to the Canola Utilization Assistance Program (CUAP), managed by the Canola Council of Canada, and were funded to operate a comprehensive check sample program. The program began in July 1980 and is currently funded to July 1983.

In this paper we shall outline the organization and goals of the program and show how the program tries to reach the goals outlined in Table I.

ORGANIZATION OF THE PROGRAM

Initially, a questionnaire was sent out to potential collaborators. The questionnaire outlined the general details of a contemplate program and asked the potential collaborators for their choice of a number of suggested ananlyses and general details about the operation of the series. Based on the response, the 4 series described in Table II were set up.

Series D originally consisted of both refined-bleached and deodorized oils but we now almost exclusively send out deodorized oils.

The program is currently a free service to collaborators but the CUAP program has requested that we start moving to a fee basis. Each collaborator subscribes to any or all series and does as many or as few analyses as he pleases. We have one large professional laboratory that does almost all analyses and several laboratories which report a single analysis. Collaborators are encouraged to employ AOCS or AOAC procedures but results are accepted from any reasonable procedure. We expected most laboratories to use AOCS or very similar AOAC procedures, since bad procedures should fall by the wayside as they fail to measure up. To a certain extent this has been the case. Several collaborators have contacted POS for advice on analysis when their results were consistently wrong.

As the series is ongoing, collaborators can join at any time. We have 35 collaborators, including oilseed crushers, government regulatory laboratories, feed testing laboratories, professional analytical laboratories, oil refining laboratories and consumer products laboratories. The common analyses usually receive 10-16 responses per month with the less popular analyses receiving 4-10 responses.

Samples are sent out at about the 7th of the month each month of the year. Results are to be returned by the 15th of the next month. POS then compiles a coded report (Table III) modelled on the AOCS reports and sends this report to all collaborators in a series.

If 6 or more responses have been received for an analysis, a trial average and standard deviation are calculated. Two standard deviations from the mean are taken as a cut-off point for good analysis results and a final average and standard deviation are calculated for all good results. As Table III shows, collaborators have a number code and receive all results for analyses in a series, with the ratio of their standard deviations from the mean to the average standard deviation for each analysis.

TABLE I

Program Goals

To provide a comprehensive check sample program to the canola industry.
To allow collaborators to evaluate analysis procedures for canola.
To provide an ongoing program for laboratories to compare their results for non-AOCS analyses, e.g., glucosinolates.
To identify test procedures which have inadequate interlaboratory precision.
To serve as the basis for collaborative studies or to introduce new test methods.
To help the canola industry standardize its test methods.

TABLE II

Series	Analyses
A (Seed)	Moisture, oil protein, glucosinolates.
B (Meal)	Moisture, oil, protein, crude fiber.
C (Crude, crude degummed oil)	Volatiles, insolubles, phosphorus, acetone insolubles, free fatty acid, neutral oil.
D (Refined deodorized oil)	Lovibond color, phosphorus, peroxide value, free fatty acid.

*Deceased, 1983.

[1]POS Pilot Plant Corporation (the initials POS denote protein, oil and starch) is a private company set up by the federal and two provincial Canadian governments and a number of food corporations to provide pilot plant services on a fee-for-use basis to clients.

TABLE III

Typical Report for Series B – Canola Meal

Analyst #	Moisture %	Oil %	Protein %	Fiber %	Deviations relative to standard deviation			
					Moisture	Oil	Protein	Fiber
1	7.05	3.21	38.63	11.4	-1.217	-0.820	0.754	-0.928
2	7.14	3.39	39.22		-0.741	0.656	1.738	
3	7.31	3.38	38.20	11.4	0.159	0.574	0.066	-0.928
4	7.30	8.75*	38.45		0.106		0.475	
5	7.25	3.41	37.46		-0.159	0.820	-1.148	
6	7.18	3.40	38.49	11.7	-0.529	0.738	0.541	-0.619
7	7.23	3.23	38.51		-0.265	-0.656	0.574	
8	7.10	3.30	38.70		-0.952	-0.082	0.885	
9	7.25	3.31	37.91	12.7	-0.159	0.000	-0.110	0.412
10	7.66	3.20	38.16		2.011	-0.902	0.000	
11	7.60	3.31	37.24	12.2	1.693	0.000	-1.508	-0.103
12	7.44	3.58	38.23	12.1	0.847	2.213	0.115	-0.206
13	6.65*	3.10	32.05*	11.6		-1.721		-0.722
14	7.47	3.18	37.68	13.7	1.005	-1.055	-0.787	1.443
15	7.19	3.33	38.56	8.9*	-0.476	0.164	0.656	
16	7.03	3.65*	36.95	14.0	-1.323		-1.984	1.753
Sample Size	15	14	15	9				
Means	7.28	3.31	38.16	12.3				
Std. Devs.	0.189	0.122	0.610	0.97				

*Result differs from the mean by greater than 2 standard deviations after first averaging and is not used in any calculations.

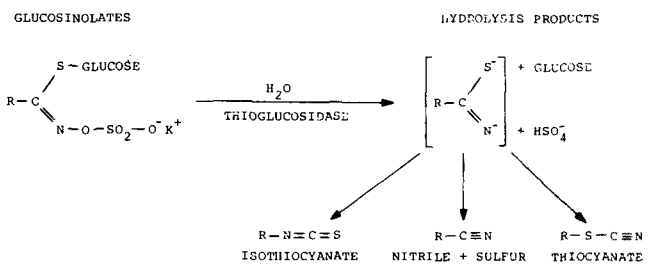

FIG. 1. Enzymatic hydrolysis of glucosinolates.

TABLE IV

Glucosinolate Procedures

Method 1	Water extraction, drying, formation of trimethylsilyl (TMS) derivatives, GLC quantitation (2).
Method 2	Water extraction, enzymatic hydrolysis, formation of thioureas, spectrophotometric quantitation (3).
Method 3	Water extraction, protein precipitation, ion exchange column clean-up, on column enzymatic desulfation, drying, formation of TMS derivatives, GLC quantiation. (4), (5).

At the end of an analysis year, collaborators are rated for each analysis for which they have a satisfactory number of responses. The rating procedure is that of the Smalley series.

Samples used in the series are solicited from collaborators, produced by POS or purchased.

General Comments on the Operation of the Program to Date

Industry response has been quite positive. Most collaborators maintain their returns faithfully for the more importamt amalyses such as oil content, protein and moisture. Responses to crude fiber, acetone insoluble matter (AIM) and neutral oil have dropped off to such an extent that we may delete these analyses.

Comments on Series A (Seed) and Series B (Meal) Analyses

It is ultimately a goal of this program to do a multidimensional analysis of variance on each of the analysis methods to sort out, where possible the effects on the analysis results of the methods employed, collaborator and the sample. At present, we can offer an analysis, based on comparing the average ratio of the standard deviation to the mean for each analysis method, as a rough method of comparing the precision of the analyses. On this basis, moisture appears to be the most imprecise of the analyses for moisture, oil and protein in Series A. For Series B, this dubious distinction is taken over by the analysis for oil content. These are, of course, relative errors. For the Series B meals, the actual standard deviation corresponds to 0.15-0.3% oil content.

Series A also contains an analysis which is unfamiliar to soybean or cottonseed processors. This is the determination of glucosinolate content. Glucosinolates, which are present in canola, cabbage, turnips, radishes and mustard have the structure shown on the left in Figure 1 and break down enzymatically to the products shown on the right. The ratio of the various breakdown products depends on the glucosinolates involved and on parameters such as pH. As stated earlier, the goals of this program include evaluating non-AOCS analyses. Table IV lists the common glucosinolate analysis procedures presently employed in Canada. Method 3, a new method, is a modification of method 1 (2) involving more extensive preparation of the sample before analysis (4,5).

Method 1 is a gas chromatographic method which involves measuring the glucosinolates as their trimethylsilyl (TMS) derivatives. Method 3 is a further modification of method 1. It involves removal of sugars and other impurities by column chromatography and removal of the sulfate group by a sulfatase before forming the TMS derivatives. Method 2 involves enzymatic hydrolysis of the glucosinolates and spectrophotometric determination of the hydrolysis products. Most of the collaborators initially used

method 1, but three employed the hydrolysis method. The latter method was often found to give low results, at the levels of glucosinolate present in canola seed, with respect to method 1. Most collaborators in the check sample program have now agreed that the hydrolysis method should be phased out for canola seed. Method 1 has shown more variation than is satisfactory among laboratories so many collaborators are contemplating using method 3, even though it is more complicated and time-consuming. At present, POS is devising a collaborative program involving the analysis by method 3 of ca. 5 samples/moth over a 3-month period. Fourteen laboratories have expressed an interest in this collaborative program. This will be the first use of the program as a basis for a collaborative study.

The other analysis in these series is crude fiber in Series B. The interest in carrying out this analysis seems to be disappearing. The problem, as we see it, is that the analysis has such a wide range of error that any result within 2 or 3% of the mean is likely to be obtained. In other words, analysis results are not much better than an educated guess. Once, in this program, a sample spiked with rapeseed hulls, low in protein and high in crude fiber, was sent out. All laboratories detected the reduction of protein content caused by the hull dilution, but only 2 out of 8 detected a higher crude fiber content. This result, we suspect, is due to the difficulty involved when the standard crude fiber analysis apparatus must deal with very fine material.

Comments on Series C Crude Oil and Series D deodorized Oil Results

In suggesting methods for the series C analyses we were guided by the outline in the Canadian standard for crude and crude degummed oil (6). This includes a requirement for a knowledge of moisture and insoluble impurities in the oil. At the beginning of this program, POS sent out quite clean and dry oils in order to avoid bacterial contamination and degumming of the oil. Thus, most of the oils had less than 0.05% moisture or insoluble impurities. In this low range, standard deviations are 20-100% of the mean. POS will try to send out wetter, more meal-contaminated samples in future if these can be adequately stabilized so that collaborators can evaluate their ability to analyze samples near legal limits of dirt and moisture.

In general, collaborators have responded well when non-normal samples have been sent out. POS has, for instance, generated 16-50 ppm phosphorus samples by cold pressing. Almost all collaborators obtained reasonable results on these low phosphorus crude oil samples without difficulty. They had no difficulty with very red refined and bleached oils but had problems with 2 extractor oils we sent out. One of these had an average of 716 ppm, with a standard deviation among collaborators of 127 ppm. It appears that some analysts were accepting results beyond the range of the linear protion of their Beer-Lambert law curves.

Acetone insoluble matter (AIM) results have standard deviations varying from 8 to 41% of the mean and averaging 22% of the mean. This analysis appears to have become less popular whereas the direct determination of phosphorus (P) has risen. The ratio of AIM/P for the first year of the study was 22.6 ± 1.7.

One of our worries in the original set-up of the program was the small number of returns for refined oil. In the last year we have managed to raise the number of collaborators in this section to 20 and thus obtain better statistics. Two analyses give problems. These are phosphorus and PV. Most of the oils POS has sent out have only a few ppm phosphorus. In the early days of the program only a few results were returned. When the program obtained enough for statistical analysis, we found that, although most labs were showing only 2-3 ppm phosphorus in the oils, some labs were finding as much as 20 ppm. Except for one lab, the discrepancy appears to be easing and we understand that this laboratory has now determined its problem. When these results are deleted, we still find that the standard deviation can be as large as the mean in the range of 2-3 ppm. Essentially, the analysis procedure is near its limit; we must find a more sensitive analysis if we wish to measure accurately the phosphorus levels in deodorized oils.

Peroxide value determination gives us some concern. At peroxide values between 0.27 and 2.33, the standard deviation of the results is 30-60% of the mean. As this method is not near its detection limit in this range, it appears that there is a problem either in preparing and shipping samples or in the methods employed in analysis.

Originally, PV results were very variable because a plastic sample bottle was used. A metal screw top enclosure can was quickly substituted and the oil was packed under nitrogen. This caused a major improvement in the precision of the analysis, especially for high PVs of 4-6. But at PV values less than one, there is usually a 50-60% deviation relative to the mean, i.e., the less stable oil with a high PV shows better precision than a better oil with a lower PV.

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