EVALUATION OF VARIOUS INDICATOR TECHNIQUES IN ESTIMATING FORAGE INTAKE AND DIGESTIBILITY BY RANGE CATTLE

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EVALUATION OF VARIOUS INDICATOR TECHNIQUES IN ESTIMATING FORAGE INTAKE AND DIGESTIBILITY BY RANGE CATTLE

AVANT PROPOS
(Preface)

Since the existence of the ruminant animal is based primarily upon its ability to utilize roughage and since a major portion of this feed is supplied by the nation's range areas, it is important to investigate the range animal's nutritional status and improve it when possible. In achieving this task one is forced toward the development of basic procedures to ascertain the quantity and quality of forage ingesta. The knowledge gained from these fundaments may provide a fulcrum from which more accurate and practical nutritional applications may be made.

Apropos to the development of basic considerations is the recognition of variables exerting influence and the adherence to experimental design to minimize this influence. Evaluation must be made of these factors and the utility of any research technique developed depends finally on this evaluation.

The use of "indicators", both external and internal, in estimating the dry matter intake and digestibility of an animal's diet has received extensive attention. The

majority of the attention, however, has been focused in areas of more confined agronomic practices, i. e., on farm type pastures where movement is restricted. These indicator techniques may offer some means of circumventing the time-consuming and onerous efforts, and the unsuitability of conventional digestibility trials for development of basic information applicable to the grazing range animal. Cognizance must be taken of the fact that these indicators are only "investigational tools" and as such possess inadequacies inherent in many biological procedures.

Embodied in the discussion to follow is a review of pertinent historical aspects of evaluation of indicators, essentially chromic oxide (Cr_2O_3), chromogen pigments and fecal nitrogen, in estimating dry matter intake and digestibility of forages by ruminants. Minor consideration is extended to the applicability of these indicators in non-ruminant species. It is superfluous to assume the responsibility for a complete treatise on the digestibility of feeds and methodology employed. The reader interested in such works is commended to reviews of the subject (Schneider, 55, p. 42; Soni, 58, p. 474-479).

In addition, procedures, results and discussion, and summary of experiments in evaluating indicator techniques in the state of Oregon are presented. The digestibility

trials and sampling were carried out at the Squaw Butte Experiment Station located in the sagebrush-bunchgrass region at Burns, Oregon and at Corvallis and the chemical analyses of herbage and fecal samples were conducted at the Department of Animal Science, Oregon State University.

In all, six animal trials were conducted. Trials 1-3 were involved with development of diurnal excretion patterns, and were carried out in the spring and summer of 1955 at Burns, Oregon. Trials 5 and 6 investigated different means of indicator administration and were done at Corvallis during the winter and spring of 1960. The last trial, testing the application of the most promising technique under range conditions, was again held at Burns in August, 1960. Details of the trials and methods of analyses are presented in a subsequent section.

TOT HOMINES QUOT SENTENTIAE
(Review of Literature)

Most of the land in the eleven western states is devoted to livestock production and range areas provide a large proportion of the roughage supply for range cattle and sheep. It is of increasing importance to expand the knowledge of better evaluating and improving this "natural resource" as a significant area in the nation's agricultural economy. In developing an improvement program for the range resource the answers to basic problems relative to the value of this feed source must be obtained. Solutions must be found to such desiderata as: The palatability of plants and their parts; quantities of various grasses and other range plants consumed by the grazing animal, the seasonal variation of plant nutrients and the evaluation of such forage as to its usefulness to the animal.

The need for obtaining basic knowledge of the grazing animal's dietary regimen has led to certain nutrition techniques to help alleviate the laborious tasks and inapplicable practices involved in digestibility studies under confined conditions. Included in the category of such techniques are the so-called "indicator methods".

Indicators may be classified into two basic divisions, e.g., the "internal" indicators and "external" indicators.

Substances belonging in the former classification are those normally found in the forage; i.e., "chromogen pigments", nitrogen and lignin. The latter group, the "external" indicators, are those which are provided to the animal from non-food sources. Examples of these are barium sulphate, monastral blue, polyethylene glycol, titanium oxide and chromic oxide. The "internal" indicators have been used as a means of estimating the apparent digestibility of various plant constituents; whereas the "external" indicators are used in estimating dry matter fecal output in lieu of taking total fecal collections.

Raymond and Minsen (52, p. 283) succinctly stated the requirements of a tracer or indicator as follows:

> "The requirements of a tracer are that it should be quantitatively recovered in the faeces (i.e., neither absorbed nor abnormally retained in the digestive tract), be non-toxic, inexpensive, readily analyzed by physical or chemical methods, and present only in small amount in the original diet."

Utilizing the two different types of indicator together; i.e., the internal indigestible plant substances to estimate herbage digestibility, and an external indicator, as chromic oxide, to estimate fecal production, estimates of herbage consumed may be attained. Misleading estimates of dry matter intake, then, may be compounded by the possible errors in the use of both these types of indicators simultaneously.

Estimation of Fecal Output Using Chromic Oxide

Chromic oxide, Cr_2O_3, satisfies the requirements of an indicator set forth by Raymond and Minson (52, p. 283) as mentioned above, with reservations. These qualifications shall be discussed later.

As mentioned previously, fecal output of grazing animals may be measured by the total collection from animals equipped with harnesses and collection bags or estimated by the administration of certain indigestible materials in known amounts to the grazing animals. The following relationship allows the estimation of fecal dry matter production:

$$\text{Dry matter fecal output in grams} = \frac{\text{Weight of external indicator fed}}{\text{Weight of indicator per gram of dry feces}}$$

The indicator approach to estimating fecal production of the grazing range animal requires a sampling procedure that will provide chromic oxide concentrations representative of a mean excretion of the indicator. The use of "grab" sampling, i.e., feces obtained directly from the rectum, has been the accepted practice by many workers (Reid et al., 54, p. 255-270; Smith and Reid, 57, p. 515-524); Hardison et al., 23, p. 11-18; 24, p. 346-352).

Such a sampling practice is not, however, without its limitation. Many workers have noted a diurnal fluctuation

in the excretion of this material. Kane et al. (32, p. 267) noted highly significant differences between the a.m. and p.m. fecal samples of dairy cows and attributed these to a "naturally occurring variation in the rate of chromium oxide excretion". They stated further that such a variation in excretion indicates that the time of day is an important consideration for the fecal sampling during a digestibility study. Smith and Reid (57, p. 5) showed an intra-day excretion of Cr_2O_3 with grazing dairy cows. Mean minimum excretion was at 2 p.m. and mean maximum excretion was at 12 midnight. Cows were administered the indicator in gelatin capsules at 7 a.m. Lancaster et al. (39, p. 117-126), using dairy cows on New Zealand dairy pastures, also observed daily fluctuation in excretion. Afternoon grab samples contained lower concentrations than adjacent morning samples. Biased estimates of fecal production were obtained using the a.m. or p.m. data singularly and were reduced by combining these data. Putnam et al. (51, p. 1723-1729) reported a "relatively smooth and symmetrical curve" of average Cr_2O_3 excretion by stall-fed Holstein cows. They stipulated that if individual observations deviated only slightly from the curve, proper correction could be made using one sampling time daily. Individual observations at any one sampling time, however, were extremely variable. Bloom et al. (3, p.

240-251) noted maximum excretions of Cr_2O_3 at 5 or 9 a.m. and minimums at 5 or 9 p.m. in dairy cows and that this pattern was largely independent of a wide variation in the physical nature of rations and feeding level. Corbett *et al.* (11, p. 266-276), working with dairy cows fed artificially dried grass, found that the concentrations of Cr_2O_3 in separate defecations varied at a maximum of ± 10 per cent from a 24-hour weighted mean concentration. Using dairy heifers fed fresh-cut alfalfa, Hardison and coworkers (24, p. 346-352) found a significant variation in the excretion of chromic oxide by sampling periods. Hardison and Reid (26, p. 35-52) also reported an irregular excretion of this reference material in grazing steers. Fecal concentrations of Cr_2O_3 varied from 55-180 per cent of the daily mean within a 24-hour period. Providing chromic oxide in a pelleted ration for Hereford heifers resulted in significant time-concentration variation of fecal Cr_2O_3 according to Elam *et al.* (19, p. 718-725). Variations appeared to be affected by feeding schedules and the magnitude of excretion precluded indiscriminate fecal sampling as regards time. Steers fed a predominantly roughage pelleted ration twice daily excreted feces that contained concentrations of Cr_2O_3 which tended to support the type of excretion patterns noted in heifers.

Bradley *et al.* (6, p. 1199) showed that chromic oxide

concentration varied from 57 to 208 per cent and from 73 to 155 per cent when administered to steers by capsule and as a part of a pelleted ration, respectively. The general shape of excretion patterns from the two dosing procedures was similar. Hardison et al. (23, p. 11-18), investigating the excretion of dairy cows found a mean maximum fecal concentration at approximately 12 noon and a mean minimum at approximately 10 p.m. Pigden and Brisson (49, p. 146-155) observed a non-uniform excretion pattern using wethers grazing experimental pastures. Shorthorn steers that grazed aftermath of a white clover-bluegrass sward were found by Brisson et al. (8, p. 90-94) to excrete this indicator in a non-uniform manner also. Japanese workers, Kameoka et al. (31, p. 462-467), reported variance in excretion of the indicator. They noted that when hay and concentrate containing Cr_2O_3 were fed at 7 and 17-hour intervals there was only one peak in excretion. When feeding was at 12-hour intervals, however, two peaks in the excretion curve were observed.

Nor is the variability of excretion of the indicator confined to ruminants. Moore (45, p. 273-288) noted differences in fecal concentration of Cr_2O_3 with pigs. Clawson et al. (9, p. 700-709) also reported concentration gradients of the indicator in pig feces. Horvath and coworkers (29, p. 869-874) found a diurnal variation in

fecal Cr_2O_3 in self-fed pigs with the concentration higher in the morning than evening. This concentration was associated with higher nitrogen and ash values in the feces. They proposed that variability in excretion of indicators was due to differences in time of digestion in the lower tract and/or differences in passage rate of different feed fractions through the stomach. Dansky and Hill (15, p. 449-460) and Mueller (46, p. 29-36) demonstrated diurnal variations in Cr_2O_3 concentration in the droppings of chickens. Moore (45, p. 286), in negating the possibilities of correlating the findings of workers studying monogastric species with those using ruminants, stated:

> "Nevertheless, it is interesting that Kane et al., (1952) found that diurnal excretion curves for Cr_2O_3 in the faeces of cows were generally opposite to those obtained for lignin, and it is, therefore, possible that the factors governing diurnal variation in the composition of the faeces of monogastric animals might also operate to some extent in ruminants."

During elucidation of the problem of variable fecal excretion of Cr_2O_3 in relation to time under a wide range of conditions, various proposals for obtaining fecal samples containing representative concentrations of chromic oxide have been afforded.

Kane et al. (32, p. 271) proposed the period from 1 p.m. to 3 p.m. as the "best time for daily sampling under the conditions of this experiment". This

time-interval offered fecal samples with Cr_2O_3 concentrations that approximated the averages of 24-hours. They also suggested another time at 4 a.m. to 6 a.m. Smith and Reid (57, p. 515-524) concluded that, despite large variation of individual samples obtained at 6 a.m. and 4 p.m., bulking fecal samples of these times resulted in concentrations representative of 24-hour collections. Hardison and Reid (26, p. 35-52) also proposed that, because of the great intra-day variation in concentrations, fecal sampling should be conducted at specified times. They also suggested the bulking of samples collected at 6 a.m. and 4 p.m. Putnam et al. (51, p. 1723-1729), reviewing the excretion curves obtained by other workers concluded that the combining of samples obtained at 12-hour intervals, regardless of sampling time, would provide fecal concentrations that were representative of the mean daily-fecal Cr_2O_3 concentration. Linkous et al. (41, p. 1009) proposed that sampling should be at 6-8 a.m. and 6-8 p.m.

Raymond and Minson (52, p. 282-296) offered a departure from the grab-sampling approach by proposing a "ring sampling" method, i.e. fecal sampling directly from the grazing sward. They emphasized that this procedure provides only an estimate of fecal production of a group of animals and not individuals. The individual variations

in grab sampling in their experiment ranged from 70-144 per cent of the mean compared to 88-120 per cent for 4 sheep over a period of 2 days.

It is apparent from the above discussion on sampling times that there is a common agreement among workers emphasizing the need of obtaining fecal samples with chromic oxide concentrations representative of mean daily concentration. However, it is also evident that there has been discord on the actual times to take samples. This conflict apparently arises from the differences in the excretion curves obtained by various workers. This divergence of opinion can be concisely summarized by a statement made by Raymond and Minson (52, p. 287), to wit:

> "These sampling methods assume there is a constant diurnal pattern of chromic oxide concentration in the faeces which is independent of the type and level of feed intake, and of the management employed. If this assumption is not valid, grab sampling will give misleading results."

Various investigators have been interested in elucidating some of the factors that affect the basic indicator excretion pattern.

Lambourne (35, p. 273-285) investigated the rate of passage of Cr_2O_3 through the digestive tract of sheep. He observed that feed quality was an important factor affecting the excretion cycle with peak concentrations

occurring progressively later on poorer pasture and on hay. A high level of feed intake (estimated), for any given feed, was often associated with an earlier appearance of the indicator in the feces, an earlier maximum of concentration and a "slightly faster fall" in Cr_2O_3 concentration. Time of administration exerted only slight effect on rate of passage, for doses given at or near times of greatest feed intake were passed more rapidly. No great amount of indicator retention was observed within the digestive tract after 72-96 hours after dosing.

Barnicoat (2, p. 202-212) reported that there was some retention of the indicator in the stomach folds of a lamb. Raymond and Minson (52, p. 282-296) presented data indicating that the pattern was affected by the level of feed intake but not by the means of administration, i.e. capsules or drench.

Corbett et al. (13, p. 343) reported on the reexamination of data wherein the abomasum of sheep was shadowed with barium sulphate to study the passage of Cr_2O_3. Using radiographs, the fate of orally administered gelatin encapsulated Cr_2O_3--a common method of dosing grazing animals with the indicator--was investigated. They found that the capsules lodged in the anterior rumen or reticulum and did not enter the omasum or abomasum. Cr_2O_3 released from the capsules was observed in the reticulum, anterior

dorsal rumen sac and the anterior region of the ventral sac. The marker was also observed "streaming through the omasal-abomasal orifice within 1 hour after dosing."

Working with dairy cows Corbett and co-workers (11, p. 266-276) concluded that the irregularity of Cr_2O_3 excretion in feces was essentially due to the uneven mixing of the indicator in the digesta of the reticulo-rumen. It was also concluded that chromic oxide was primarily associated with the solid fraction of the digesta.

Brisson et al. (8, p. 93) offered the following comment:

> "It appears, therefore, that an important factor controlling the shape and amplitude of the Cr_2O_3 excretion curve of grazing animals is the relationship between time of chromic oxide administration and grazing periods.
>
> It is common knowledge that changes in weather, grazing management and other factors can markedly influence the grazing behaviour of ruminant animals. These factors, then, would modify the shape of the Cr_2O_3 excretion curve, the time of dosing remaining constant."

Moore (45, p. 284), in attempting to resolve this problem with pigs, stated that the differential passage of Cr_2O_3 might be caused indirectly by a variable rate of passage of the food constituents with differing digestibility. Feces derived from food material with high digestibility theoretically would contain higher concentrations of Cr_2O_3 while those originating from less digestible food

fractions would contain lower concentrations.

Balch and co-workers (1, p. 184-197), studying factors influencing the rate of excretion of Cr_2O_3 by steers found a rapid excretion of the indicator into the omasum during the first 30-60 minutes after administration. Posture, i.e. standing or lying, did not exert any marked effect on Cr_2O_3 concentration in the feces. Dosing animals with the indicator immediately prior to a single daily feed resulted in a more even pattern of fecal Cr_2O_3 excretion than administration after feeding. They recommended that Cr_2O_3 used in grazing trials be given immediately preceding the daily grazing period and that the suitability of any program of Cr_2O_3 administration and fecal sampling be checked against the total fecal collection of each experiment.

The discovery that the indicator was excreted in an irregular fashion perpetrated investigations in means of reducing the uneven excretion and ultimately in increasing the accuracy of estimates of fecal production. Raymond and Minson (52, p. 282-296) found no reduction in the diurnal excretion by using a drench of Cr_2O_3 when compared to encapsulated Cr_2O_3 in sheep studies. Mahaffey *et al.* (43, p. 672) noted a reduction in the diurnal cycle of steers when a Cr_2O_3-collodion mix was fed compared to three other methods of dosing. Hardison *et al.* (23, p.

11-18) reported that the variability in Cr_2O_3 excretion and random error was reduced by providing the indicator twice daily to cows instead of once daily. No difference was noted between "at-feeding" or "between-feeding" administration of the indicator. Brisson and co-workers (8, p. 90-94) noted significant changes in Cr_2O_3 concentration when steers were dosed once or twice daily. Providing the indicator 6 times a day resulted in a constant excretion rate. Pigden and Brisson (49, p. 146-155) noted distinctly different diurnal excretion patterns between 2 grazing wethers that were dosed once daily with Cr_2O_3. No diurnal pattern was noted in wethers dosed 6 times a day.

Pigden and Brisson (50, p. 185) reported on the use of a "sustained-release pellet" composed of Ca_2SO_4 and Cr_2O_3 as a means of reducing the amplitude of excretion of the indicator. The use of paper as a carrier of chromic oxide has been reported by Corbett et al. (12, p. 1014-1016) as a method to provide a consistent excretion rate. The same workers used this procedure with sheep and found a reduction in variation of fecal Cr_2O_3 concentration compared to gelatin encapsulated material.

The feasibility of Cr_2O_3 as a means of measuring grazing animal fecal production is dependent upon a quantitative recovery of the material. This recovery does not need to be 100% but a major requisite is that recovery

should be consistent within and among trials. If this is the case, proper adjustments may be made. The problem of percentage recovery of Cr_2O_3 is directly related to the adequacy of chemical analysis and to the diurnal excretion and its causes, but a brief discussion should be given to this area since certain workers have obtained variable results without investigating the excretion pattern per se.

Crampton and Lloyd (14, p. 319-326) noted average recoveries of the indicator of 98.5 per cent when the Cr_2O_3 was mixed with grain for sheep but recoveries of 86 per cent when it was provided as a capsule. Putnam and co-workers (51, p. 1723-1729) found a recovery of 99.6 per cent with cows fed roughage. Raymond and Minson (52, p. 286) noted a mean recovery of 96.9 per cent with sheep dosed with a Cr_2O_3-drench and 99.96 per cent with sheep receiving capsules containing the indicator. Hardison et al. (24, p. 346-352) observed that the recovery of Cr_2O_3 decreased as the dosage to dairy animals increased. Animals receiving a 5 gram dose daily had a recovery of 101-104 per cent while those on a 45 gram dose had a recovery from 81 to 83 per cent. Moore (45, p. 273-288) reported a mean recovery of 83 per cent with pigs while Clawson et al. (9, p. 700-709), also working with pigs, noted relative Cr_2O_3 percentage recoveries ranging from 97.9 to

103.3 per cent depending upon sampling times. The mean rate of Cr_2O_3 recovered in feces of dairy cows was found by Smith and Reid (57, p. 515-524) to be 100.6 per cent when they combined fecal samples taken at 6 a.m. and 4 p.m. Pigden and Brisson (49, p. 146-155) reported recovery values with sheep from 101 per cent and 94 per cent in two trials to 87 per cent in a third trial.

Balch and co-workers in 1957 (1, p. 196) summarized the difficulties of using chromic oxide as a means of measuring fecal production as follows:

> "It appears unlikely that a method of giving Cr_2O_3 to grazing animals will be found that ensures an even excretion in the faeces and, although under conditions studied previously, sampling at 05:00-06:00 hrs. and 13:00-16:00 hrs. gave satisfactory results it would be unwise to assume that it will necessarily be so in all experiments. It is therefore recommended that the suitability of the sampling times should be checked against complete collection of faeces under the conditions of each experiment. This would seem to be especially necessary where unusual crops or unusual grazing management are being used."

Evidence cited represents a cacophony of opinions for the use of the indicator. Its applicability for range grazing trials can only be ascertained by actual experimentation, but one needs to accept it with caution as a panacea for all the ills befalling studies of digestibility and intake of range forage.

Estimation of Apparent Digestibility Using "Chromogen Pigments" and Fecal Nitrogen

Paralleling the problems in measuring fecal production of the grazing range animal is the difficulty in assessing the digestibility of range forage. A further and greater limitation in range grazing trials is that there is no accurate standard means of determining digestibility coefficients against which experimentally deduced values may be compared. This disturbance may not be of great consequence in pasture trials where sward sampling and/or hand-feeding comparable animals pasture herbage may allow reasonable estimates of digestibility, but it is a larger task in range trials where clipped forage for hand feeding may or may not reasonably duplicate that consumed by the grazing animal. The situation is further complicated where animals graze native range areas containing 5 or more dominant grasses indigenous to a particular area.

Schneider, Soni, and Ham (55, p. 1-42) presented a comprehensive review of the literature relative to the methods of determining consumption and digestibility of forages, with pertinent reviews on the applicability of various indicator techniques. No attention will be afforded to forage clipping and sampling as a means of estimating intake and digestibility in this thesis, since these practices are outside the sphere of indicator techniques. This, however, does not negate their potential

in this problem.

The use of "chromogen pigments" as an indicator of digestibility has received widespread attention since the original reports of Reid et al. (53, p. 60-71; 54, p. 255-270). In their first publication these workers proposed that plant pigments or "chromogenic substances" might serve as indigestible internal indicators to estimate forage digestibility. Data presented showed good agreement between digestibility coefficients obtained in this manner and those by conventional methods, under pasture conditions. Recognizing the problem of adequate sward-sampling to obtain herbage comparable to that consumed by grazing animals, these workers in the latter report offered a regression equation, whereby forage chromogen could be calculated by known fecal chromogen concentration. Varying results have been obtained using this indicator. In general, however, results have indicated its acceptability under most stall-feeding and succulent pasture conditions using sheep, cows and steers (Reid et al., 53, p. 60-71, 54, p. 255-270; Hardison and Reid, 26, p. 35-52; Schneider, Soni, and Ham, 55, p. 1-42; Brisson, Angus and Sylvestre, 7, p. 528-532; Bradley et al., 5, p. 1279; and others).

Cook and Harris (10, p. 565), on the other hand, stipulated that the chromogen method was not suitable for

determining digestibility of winter range forage for sheep. Brisson and co-workers (7, p. 528-532) remarked upon the acceptability of the procedure, but stated that further investigations should be conducted to ascertain its applicability under various grazing conditions. Kane and Moore (33, p. 936), in applying the formula of Reid et al. (54, p. 255-270), found that there was an average error of 16 per cent between calculated digestibility values and conventional methods. They recommended that a correction be applied to the formula for seasonal forage differences due to locality. Kennedy et al. (34, p. 627-638) found this formula to be generally unsatisfactory in estimating dry matter digestibility from fecal pigment concentrations.

Weswig et al. (60, p. LV) using chromogen pigments in evaluating grass genotypes, found that digestibility coefficients determined in this manner did not consistently duplicate those obtained conventionally in rat experiments. Petersen et al. (48, p. LVII), using the formula given by Reid et al. (54, p. 255-270), proposed the determination of fecal chromogen from known forage chromogen values. They found that an explicit mathematical solution was not possible for fecal chromogen in terms of forage chromogen because of the logarithmic nature of this relationship. Also, when digestibility coefficients obtained by this method were plotted as a function of forage chromogen they

passed through a minimum of 80 units of chromogen per gram of dry matter. The same digestibility could then be predicted for two forages of different chromogen content. They reported that the following equation provided estimates of dry matter digestibility that agreed favorably with those obtained in conventional trials with sheep.

$$\text{Per cent Digestibility} = 96.6491 - \frac{310.0510}{\sqrt{\text{Forage chromogen}}}$$

where forage chromogen is given in units chromogen per gram dry matter, as defined by Reid *et al.* (54, p. 255-270).

This equation, at first glance, would seem to be inappropriate since adequate forage sampling is an important factor. If, however, recent techniques of animal sampling could be used, *vide*, Lesperance *et al.* (40, p. 682-689), employing the esophageal fistula, and the preliminary rumen clearance investigations at the Oregon station, a proposal such as this might have great merit.

Nelms *et al.* (47, p. 123-128), working with beef cattle, showed a diurnal excretion pattern of these pigments which was in contrast to preceding published results. Hardison and co-workers (25, p. 768-773) substantiated this and stated that the excretion pattern for these pigments was of the general form as that reported by some workers for chromic oxide although the variation was

considerably less. They noted that digestibility estimates from fecal samples taken at 6 p.m. were significantly lower than coefficients calculated from total fecal collections. Coefficients calculated from feces at other times did not differ significantly from those by conventional methods. Soni et al. (58, p. 474-479) reported no repeatable diurnal pattern of digestibility coefficients calculated from fecal chromogen concentrations in grazing trials with sheep and cattle. Lancaster and Bartrum (38, p. 489-496) emphasized the need for extreme care in analytical procedures in determining the chromogen pigment concentrations and concurrence was given by Schneider, Soni and Ham (55, p. 1-42).

Irwin et al. (30, p. 541-551) discussed the role of plant pigments in estimating digestibility and concluded that certain pigments of the complex were too unstable and too digestible to be used as indicators of digestibility. In 1957 Mixner et al. (44, p. 67-74) found at least six individual chromogen pigments in dairy cow urine and that urinary excretions of these pigments followed a distinct diurnal pattern. Davidson (16, p. 5; 17, p. 86-92; 18, p. 209-212), from studying the fate of major plant pigments in the rumen and alimentary tract of sheep and the relation of these "chromogens" to one another, concluded that it would be inadvisable to apply the chromogen method widely

until comparative digestibility trials have shown its reliability with other collection procedures. Smart, Matrone and Smart (56, p. 1331-1332) found that the chromogen ratio method was unsatisfactory in determining digestion coefficients of switch cane, a coarse, high-fiber roughage somewhat similar to range forage.

In 1949 Lancaster (36, p. 31-38) reported a method of determining the digestibility of forage based on the nitrogen content of feces of sheep grazing New Zealand pastures. He reported comparisons of digestibility coefficients of organic matter calculated in this manner with those obtained from 52 digestibility trials. He proposed that nitrogen excreted in sheep feces per unit intake of pasture organic matter is a constant, C. This value from his experience was 0.83 ± 0.102. Gallup and Briggs (22, p. 110-116), however, indicated that the ratio of fecal nitrogen to dry matter consumed increased slightly with increasing protein content. Homb and Breirem (28, p. 496-500) also reported an increase in this ratio and Forbes (20, p. 19-23) showed that there was a steady increase in fecal N per 100 grams of dry matter intake as the feed protein increased. Data on fresh grass were not adequate to warrant definite conclusions other than the fact that fecal N varied widely. In 1950 Forbes (21, p. 231) reported that the "protein digestibility" may be used with

a satisfactory degree of accuracy for determination of dry matter digestibility of grazing steers. Subsequently, Lancaster (37, p. 15-20) presented a modification of the earlier method and its applicability with cattle grazing New Zealand pastures. This utilized a regression equation to express the increased feed:feces nitrogen ratio. The range in digestibility of pasture studied was 58-84 per cent. Soni et al. (58, p. 474-479) reported that there was no diurnal variation in the estimates of digestibility using fecal N and that the method appeared to give as good an estimate of dry matter digestibility as the chromogen method. Kennedy and co-workers (34, p. 627-638) observed that fecal nitrogen provided a more reliable index of digestibility than did the chromogen method.

The preceding discussion on the use of chromic oxide, chromogen pigments and fecal nitrogen as indicators in estimating fecal output and digestibility demonstrates, to a small degree, the complications involved in assessing dry matter intake of grazing animals. The investigations reported herein were an endeavor to extend some of the successes obtained elsewhere with animals fed succulent type feeds to grazing and stall-fed steers consuming forages indigenous to the sagebrush-bunchgrass region.

SUI GENERIS
(Materials and Methods)

The indicator techniques under scrutiny in the first part of this investigation were those involving the "chromogen pigments", fecal nitrogen: dry matter ratio and chromic oxide; the first two as a means of predicting the apparent dry matter digestibility and the latter as a means of estimating the amount of dry matter excreted via the feces.

Trials 1, 2 and 3

Six grade Hereford steers were used in each of three digestibility trials conducted on improved range where the forage was predominantly crested wheatgrass, Agropyron desertorum, at three stages of maturity; i.e., immature forage-June, mature forage-July and dried mature forage-September. Each trial consisted of a five-day preliminary period and a five-day collection period. In the interim between trials the steers grazed crested wheatgrass range immediately adjacent to the small test areas. Three steers were hand-fed clipped crested wheatgrass herbage during each trial and three grazed one-acre pastures. The electrically fenced pastures were of a size that forced the grazing animal to graze the herbage close to the ground, therefore the clipped herbage was obtained by

harvesting as close to the ground as practical for the sake of similarity. The stall-fed animals received the herbage ad lib. with the rations renewed twice daily, at 7:30 a.m. and 3:30 p.m. Water was provided to the confined steers prior to feeding with ample time allowed for fill. In the case of the grazing steers water was available at all times.

Five grams of chromium oxide were administered once daily to each steer via 10-oz. gelatin capsules administered by balling gun at 7:00 a.m. beginning the first day of the preliminary period and ending the last day of the collection period.

Total fecal collections were obtained from each steer by collection bags, changed twice daily at 6 a.m. and 6 p.m. Ten per cent aliquots of the excreta were taken to represent composite fecal samples for the five days for each steer. In addition, "grab" fecal samples were taken at 2-hour intervals from each steer for the five days of each trial. All fecal samples were stored in 6 mil. plastic bags at 0-5°C. for chemical analyses.

Analyses included determinations of "chromogen pigments" (Reid, et al., 53, p. 60-71), dry matter percentage, crude protein percentage, and chromic oxide concentration (Bolin et al., 4, p. 634-635). In addition, proximate analyses were made on the composite fecal samples from

each steer on each trial. Clipped herbage, as well as pasture herbage, was sub-sampled for proximate analyses and determinations of "chromogen pigments". The analytical results were punched onto IBM cards to allow the use of data processing equipment and expedite statistical analysis.

Trials 5 and 6

These two trials were initiated to investigate further the value of the external indicator, chromic oxide, as a means of accurately estimating fecal dry matter output of range cattle when fed forage indigenous to the range area. Trial 5 was designed to obtain information on the applicability of various methods of administering Cr_2O_3 in accurately estimating dry matter output and trial 6 was initiated to investigate to what extent limited feed intake affected the reliability of estimates using Cr_2O_3 incorporated on cellulose fibers.

Six grade Hereford steers, approximately 12 months of age, were used in the two trials. The steers were chosen on the basis of uniformity in weight and age from the herd of the Squaw Butte Experiment Station. They were housed in an open barn and individually fed twice daily on native flood-meadow, rush-sedge hay from the range area.

In trial 5 a comparison was made of three different

means of providing Cr_2O_3 wherein two steers were allocated randomly to each of three treatments: (1) chromic oxide as a powder administered in gelatin capsules, (2) chromic oxide as a powder administered in cottonseed meal in the feed, and (3) chromic oxide dispersed on purified cellulose fibers (Solka-Floc[1]) and also given in gelatin capsules. All animals were fed ad libitum twice daily at 8 a.m. and 4 p.m. The steers were fitted with fecal collection bags and total fecal collections were made for 5 days following a 5-day preliminary period. Cr_2O_3 was administered to all steers during the preliminary and collection periods. "Grab" fecal samples were taken twice daily at 8 a.m. and 4 p.m. for the 5-day trial. The steers in groups 1 and 2 received 10 g. chromic oxide per day in two 5 g. doses. The animals in group 3 received approximately 10 g. per day in two doses but it was not possible to maintain exact amounts between days and animals due to the manner in which the mix was prepared. Exact amounts of Cr_2O_3 given this group were calculated from weight and analysis of the mixture administered. In all groups chromic oxide was administered at 8 a.m. and 4 p.m.

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[1] Solka-Floc is a purified wood cellulose product of the Brown Co., Berlin, New Hampshire.

Trial 6 involved a cross-over design to study the effect of limited feed intake on the reliability of the external indicator in estimating total fecal output. Three steers were randomly assigned to each of two groups: ad libitum and limited hay consumption. Nature of feed, feeding and indicator administration times, lengths of preliminary and collection periods, and the procedure of fecal collections were similar to those described for trial 5. Each steer received approximately 10 g. of Cr_2O_3 dispersed on cellulose in two daily doses.

The Cr_2O_3-cellulose mix was prepared in large enough quantities to last the duration of one trial. The mix consisted of about 60 per cent Cr_2O_3, 39 per cent Solka-Floc and 1 per cent Al_2SO_4. The Al_2SO_4 was used as a mordant to insure adherence of the chromic oxide onto the cellulose fibers (Corbett et al., 12, p. 1014-1016). Following thorough mixing and drying, the mix was packed into weighed and numbered no. 10 gelatin capsules. The capsules were reweighed and the amount of mix per capsule was computed. Chromic oxide concentration was determined by chemical analysis of the mix (Bolin et al., 4, p. 634-635). Each steer received designated capsules thus allowing computation of exact amounts of chromic oxide given to each animal.

Total dry matter content and chromic oxide concentration (Bolin et al., 4, p. 634-635) were determined on all fecal samples. Data were analyzed statistically, according to the method of Lucas (42).

NATURA NON FACIT SALTUM
(Results)

Evaluation of Chromic Oxide

In considering Cr_2O_3 as an indicator for measuring the fecal output of grazing range animals one needs to determine its relative recovery under controlled conditions. Such data are presented in Table 1 for trials 1, 2, and 3. The range in recovery was 94 to 126 per cent for animals on both regimes in trial 1, while the mean recovery for the grazing and hand-fed animals was 121 and 102 per cent, respectively, for this trial. In trial 2 the range in recovery was 99 to 114 per cent and the mean recovery was 109 and 101 per cent for the grazing and hand-fed animals, respectively. Percentage recovery of chromic oxide in trial 3 was above 100 with all six steers and ranged from 112 to 138 with a mean recovery of 132 for the grazing steers and 123 for the hand-fed animals. Recovery of this indicator was not consistent among animals, i.e., the steers with the lowest recoveries in the first trial were not consistently low in the succeeding trials. However, those steers that were confined had mean percentage recoveries that were lower than those of the grazing animals in all three trials and, in two of these trials the recovery percentages of the hand-fed animals were near 100 per cent (vide, 102 and 101) respectively for trials

1 and 2.

The collection of rectal fecal samples every 2 hours for 5 days from all steers allowed an investigation of the excretion pattern of chromic oxide during each of the three trials. Figure 1 shows the mean chromic oxide concentration of the three grazing steers and hand-fed steers in the three trials. The concentration in the "grab" samples exhibited a definite diurnal pattern, however this pattern varied somewhat between animals on either regime within any one trial. Greater discrepancies between the excretion curves of various trials were also noted. Average maximum and minimum Cr_2O_3 concentrations, time of occurrence and mean concentration for each trial and on each regime within trials are summarized in Table 2.

The ultimate acceptability of Cr_2O_3 as an investigational tool is dependent on accuracy in estimating values for which it is employed. The fecal production of grazing range steers needs to be estimated accurately and consistently under a wide range of conditions by this procedure, or its usefulness may be diminished to the point of ineffectiveness.

Average dry matter fecal outputs measured by total collections and estimated by mean concentration of Cr_2O_3 in: (1) composite fecal samples of each steer, (2) all

Table 1. Per cent of administered chromic oxide recovered in feces from steers grazing or hand-fed crested wheatgrass.[1]

Trial No.	Animal No.	Group	% Cr_2O_3 recovered
1	1	grazing	126
immature	3		124
herbage	5		112
	Mean		121
	2	hand-fed	103
	4		109
	6		94
	Mean		102
2	1	grazing	102
mature	3		112
herbage	5		114
	Mean		109
	2	hand-fed	102
	4		99
	6		102
	Mean		101
3	1	grazing	138
mature-dry	3		132
herbage	5		127
	Mean		132
	2	hand-fed	130
	4		128
	6		112
	Mean		123

[1] 5 grams Cr_2O_3 in 10 g. gelatin capsule given once daily for 5 days.

Figure 1. Average Chromic Oxide Excretion of Steers Grazing or Hand-Fed Crested Wheatgrass

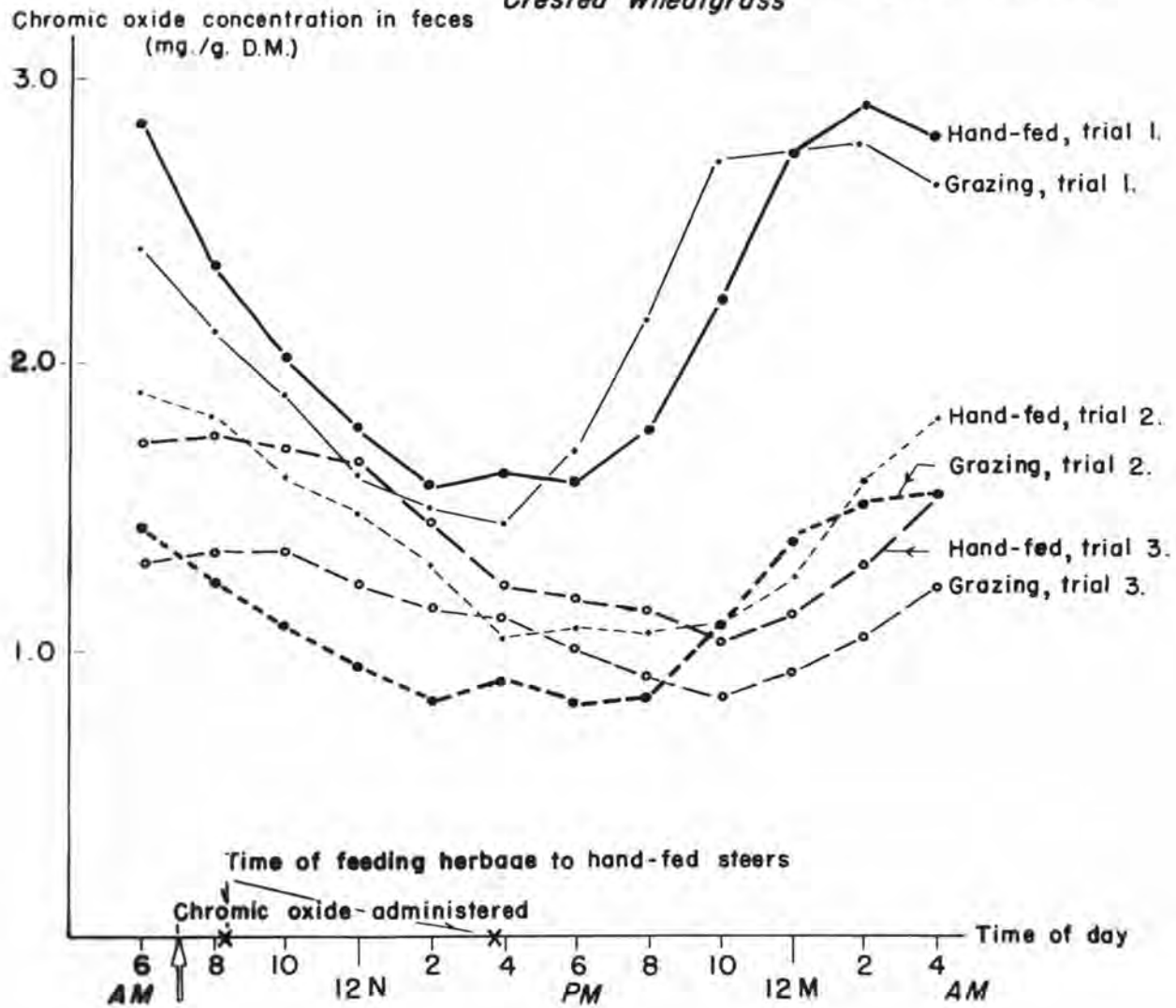


Table 2. Average maximum and minimum Cr_2O_3 concentrations in dry matter of "grab" fecal samples of steers hand-fed and grazing crested wheatgrass.

	Trial 1		Trial 2		Trial 3	
	H.F.	Grazing	H.F.	Grazing	H.F.	Grazing
	mg/g	mg/g	mg/g	mg/g	mg/g	mg/g
Maximum	2.94	2.79	1.91	1.56	1.74	1.36
Time	2 am	2 am	6 am	4 am	8 am	10 am
Minimum	1.57	1.46	1.08	0.82	1.04	0.84
Time	2 pm	4 pm	8 pm	6 pm	10 pm	10 pm
Mean	2.19	2.15	1.45	1.12	1.38	1.12

"grab" samples and (3) "grab" samples obtained at 6 a.m. and 4 p.m. are presented in Tables 3, 4 and 5 for steers grazing and hand-fed crested wheatgrass during trials 1, 2 and 3, respectively. These data are rearranged on the basis of each method of calculation for the three trials in order to facilitate the comparisons of one method under three different stages of herbage maturity and are presented in Tables 6, 7 and 8.

In trial 1 estimates of fecal production for steers on both regimes determined by the three indicator procedures varied from 20 per cent underestimation to 49 per cent overestimation. Average Cr_2O_3 concentrations in the composite fecal samples of total collections consistently underestimated the actual fecal production of steers on both regimens. There was no consistency in estimates using the other two procedures (Table 3).

When steers consumed more mature forage, estimates of fecal output ranged from -22 to +47 per cent of actual measured amounts (Table 4). As in trial 1, calculation from composite sample concentrations consistently underestimated fecal production.

The range in estimates using these three procedures was from 34 per cent underestimation to 54 per cent overestimation in trial 3. Composite samples again underestimated fecal output. There was a wide range of

Table 3. Average dry matter fecal output (DMFO), lb. per day, measured by total collections and estimated using Cr_2O_3 by three different means for steers hand-fed and grazing immature crested wheatgrass.

	Trial 1							
	Hand-fed Steers				Grazing Steers			
	2	4	6	Mean	1	3	5	Mean
Actual D.M.F.O.	4.5	4.8	4.5	4.6	6.0	6.6	5.7	6.1
Estimated F.O.[1]	4.3	4.2	4.3	4.3	4.8	5.4	5.0	5.1
% Difference	-4.4	-12.5	-4.4	-6.5	-20.0	-18.2	-12.3	-16.4
Estimated F.O.[2]	4.6	4.8	5.9	5.0	4.8	5.3	5.4	5.1
% Difference	2.2	0.0	31.1	8.7	-20.0	-19.7	-5.3	-16.4
Estimated F.O.[3]	4.6	4.7	5.7	5.0	5.4	5.8	6.0	5.7
% Difference	2.2	-2.1	26.7	8.0	-10.0	-12.1	5.3	-6.6

1 Estimated from composite samples.
2 Estimated from mean of all "grab" samples.
3 Estimated from bulking "grab" samples taken at 6 am and 4 pm.

Table 4. Average dry matter fecal output (DMFO), lb. per day, as measured by total collections and estimated using Cr_2O_3 by three different means for steers hand-fed and grazing mature crested wheatgrass.

	Trial 2							
	Hand-fed Steers				Grazing Steers			
	2	4	6	Mean	1	3	5	Mean
Actual D.M.F.O.	7.0	7.3	6.0	6.8	8.6	8.7	9.1	8.8
Estimated F.O.[1]	6.2	6.1	5.7	6.0	7.8	6.9	7.1	7.3
% Difference	-11.4	-16.4	-5.0	-11.8	-9.3	-20.7	-22.0	-17.0
Estimated F.O.[2]	7.7	10.7	5.8	7.6	8.7	10.6	10.1	9.8
% Difference	10.0	46.6	-3.3	11.8	1.2	21.8	11.0	11.4
Estimated F.O.[3]	7.0	9.5	5.8	7.4	8.9	9.5	10.8	9.7
% Difference	0.0	30.1	-3.3	8.8	2.3	20.7	18.7	10.2

1 Estimated from composite samples.
2 Estimated from mean of all "grab" samples.
3 Estimated from bulking "grab" samples taken at 6 a.m. and 4 p.m.

Table 5. Average dry matter fecal output (DMFO), lb. per day, as measured by total collections and estimated using Cr_2O_3 by three different means for steers hand-fed and grazing dry-mature wheatgrass.

	Trial 3							
	Hand-fed Steers				Grazing Steers			
	2	4	6	Mean	1	3	5	Mean
Actual D.M.F.O.	7.6	7.9	6.7	7.4	8.4	9.7	9.3	9.1
Estimated F.O.[1]	5.3	5.5	5.0	5.3	5.6	7.1	6.1	6.3
% Difference	-30.3	-30.4	-25.4	-28.4	-33.3	-26.8	-34.4	-30.8
Estimated F.O.[2]	7.4	7.9	8.7	8.0	8.1	8.6	14.3	9.8
% Difference	-2.6	0.0	29.8	8.1	-3.6	-11.3	53.8	7.7
Estimated F.O.[3]	7.2	7.6	7.5	7.4	7.3	8.2	12.8	9.4
% Difference	-5.3	-3.8	11.9	0.0	-13.1	-15.5	37.6	3.3

1 Estimated from composite samples.
2 Estimated from mean of all "grab" samples.
3 Estimated from bulking "grab" samples taken at 6 a.m. and 4 p.m.

Table 6. Average dry matter fecal output as measured by total collections and estimated by Cr_2O_3 in composite fecal samples from steers hand-fed or grazing crested wheatgrass.

Trial No.	Steer No.	Group	Dry matter fecal output		
			Actual lb./day	Estimated lb./day	% Difference from Actual
1	1	grazing	6.0	4.8	-20.0
	3		6.6	5.4	-18.2
	5		5.7	5.0	-12.3
	Mean		6.1	5.1	-16.4
	2	hand-fed	4.5	4.3	-4.4
	4		4.8	4.2	-12.5
	6		4.5	4.3	-4.4
	Mean		4.6	4.3	-6.5
2	1	grazing	8.6	7.8	-9.3
	3		8.7	6.9	-20.7
	5		9.1	7.1	-22.0
	Mean		8.8	7.3	-17.0
	2	hand-fed	7.0	6.2	-11.4
	4		7.3	6.1	-16.4
	6		6.0	5.7	-5.0
	Mean		6.8	6.0	-11.8
3	1	grazing	8.4	5.6	-33.3
	3		9.7	7.1	-26.8
	5		9.3	6.1	-34.4
	Mean		9.1	6.3	-30.8
	2	hand-fed	7.6	5.3	-30.3
	4		7.9	5.5	-30.4
	6		6.7	5.0	-25.4
	Mean		7.4	5.3	-28.4

Table 7. Average dry matter fecal output as measured by total collections and estimated by Cr_2O_3 in mean "grab" fecal samples from steers hand-fed or grazing crested wheatgrass.

Trial No.	Steer No.	Group	Dry matter fecal output		
			Actual lb./day	Estimated lb./day	% Difference from Actual
1	1	grazing	6.0	4.8	-20.0
	3		6.6	5.3	-19.7
	5		5.7	5.4	-5.3
	Mean		6.1	5.1	-16.4
	2	hand-fed	4.5	4.6	2.2
	4		4.8	4.8	0.0
	6		4.5	5.9	31.1
	Mean		4.6	5.0	8.7
2	1	grazing	8.6	8.7	1.2
	3		8.7	10.6	21.8
	5		9.1	10.1	11.0
	Mean		8.8	9.8	11.4
	2	hand-fed	7.0	7.7	10.0
	4		7.3	10.7	46.6
	6		6.0	5.8	-3.3
	Mean		6.8	7.6	11.8
3	1	grazing	8.4	8.1	-3.6
	3		9.7	8.6	-11.3
	5		9.3	14.3	+53.8
	Mean		9.1	9.8	7.7
	2	hand-fed	7.6	7.4	-2.6
	4		7.9	7.9	0.0
	6		6.7	8.7	+29.8
	Mean		7.4	8.0	8.1

Table 8. Average dry matter fecal output as measured by total collections and estimated by Cr_2O_3 in "grab" samples collected at 6 a.m. and 4 p.m. from steers hand-fed or grazing crested wheatgrass.

Trial No.	Steer No.	Group	Dry matter fecal output		
			Actual lb./day	Estimated lb./day	% Difference from Actual
1	1	grazing	6.0	5.4	-10.0
	3		6.6	5.8	-12.1
	5		5.7	6.0	+5.3
	Mean		6.1	5.7	-6.6
	2	hand-fed	4.5	4.6	+2.2
	4		4.8	4.7	-2.1
	6		4.5	5.7	+26.7
	Mean		4.6	5.0	+8.7
2	1	grazing	8.6	8.9	+2.3
	3		8.7	9.5	+20.7
	5		9.1	10.8	+18.7
	Mean		8.8	9.7	+10.2
	2	hand-fed	7.0	7.0	0.0
	4		7.3	9.5	+30.1
	6		6.0	5.8	-3.3
	Mean		6.8	7.4	+8.8
3	1	grazing	8.4	7.3	-13.1
	3		9.7	8.2	-15.5
	5		9.3	12.8	+37.6
	Mean		9.1	9.4	+3.3
	2	hand-fed	7.6	7.2	-5.3
	4		7.9	7.6	-3.8
	6		6.7	7.5	+11.9
	Mean		7.4	7.4	0.0

estimates using mean concentrations of all "grab" samples, or when "grab" samples collected at 6 a.m. and 4 p.m. were combined.

The composite concentrations represent an average concentration of all feces voided into the collection bags and should give accurate estimates of fecal production. If the estimates are corrected for percentage recovery of the indicator, closer agreement to the actual feces produced is achieved but there is still some disagreement. This correction, however, would necessitate a calculation of recovery for each set of conditions under which the indicator is applied. Average concentrations of all "grab" samples obtained from each steer are computed from approximately 60 samples and should represent a mean excretion of Cr_2O_3. There was, however, extreme variability in fecal estimates using this procedure among steers and trial conditions. Determining fecal production estimates as recommended by previous workers; i.e., using sampling times of 6 a.m. and 4 p.m., did not result in any more consistent estimates. There was no apparent uniformity in estimates between the three grazing steers and those hand-fed using the three estimating procedures.

Since the first three trials were conducted, wherein chromic oxide was administered once daily, additional information has appeared in the literature, as mentioned

previously. Results using confined and pasture-grazed animals indicated that additional times of dosing animals with the indicator, as well as other dosing procedures, resulted in a less variable excretion and increased the plausibility of utilizing selected fecal sampling times to estimate fecal production of grazing animals. Following trials took cognizance of these findings.

The results of trial 5, involving three methods of indicator administration at 8 a.m. and 4 p.m., with fecal sampling at the same times, are presented in Table 9. Actual dry matter output was quite variable within and among groups. Fecal output was more accurately estimated using the indicator either in cottonseed meal or as a mix with cellulose than as the pure salt; however the average estimate in both cases was 0.8 lb. per day above the actual dry matter production. Estimates were 119 and 123 per cent of actual for cottonseed meal and cellulose as carriers, respectively. Average per cent recovery of the indicator for the three groups was: cottonseed meal as a carrier, 84; "free" Cr_2O_3 in capsules, 73; and Cr_2O_3-cellulose in capsules, 81, respectively.

The influence of amount of hay intake on the reliability of a Cr_2O_3-cellulose mix in estimating fecal production is summarized in Table 10. Average dry matter output was estimated within 0.8 and 0.7 lb. per day with

Table 9. Comparison of dry matter fecal output measured by collection bags and estimated by Cr_2O_3

Means of Administration	Steer No.	Fecal Output (lb. D.M./day) Actual	Estimated	Estimated at % of Actual
Cottonseed Meal	13	4.0	5.0	124
	16	4.7	5.4	114
	mean	4.35	5.20	119
"Free" Cr_2O_3 in capsules	14	3.7	4.7	126
	15	2.8	4.2	150
	mean	3.25	4.45	138
Cr_2O_3-Solka-Floc in capsules	17	3.7	4.7	127
	18	3.2	3.8	119
	mean	3.45	4.25	123

Table 10. Effects of hay intake on estimates of dry matter fecal output by Cr_2O_3 in a cellulose carrier and total collections. Cross-over trial employed.

Treatment	Mean Measured D.M.F.O. (lb./day)	Mean Estimated D.M.F.O. (lb./day)	Recovery Cr_2O_3 (%)
Ad. libitum	4.36	5.18*	84.2
Limited	4.08	4.79	85.4
LSD 5%	0.45	0.30	6.8
1%	0.61	0.41	9.2

* Significantly different ($P < 0.05$) from estimated output of steers on limited intake.

steers on a full-feed of hay and on limited hay, respectively. The amounts of administered Cr_2O_3 recovered were 84 and 85 per cent, respectively, for the full-fed and limited fed groups.

Dry Matter Digestibility Estimated From Chromogen Pigments

In preliminary studies in range trials it was of interest to determine the recovery of chromogen pigments, especially since no previous information existed with regard to cattle consuming range herbage. In Table 11 the percentage recovery of these pigments are presented for each hand-fed steer for trials 1, 2 and 3. Average recoveries were highly variable between trials ranging from 67 to 109 per cent. The mean percentage recovery was 72, 68 and 97 for the three steers in trials 1, 2 and 3, respectively.

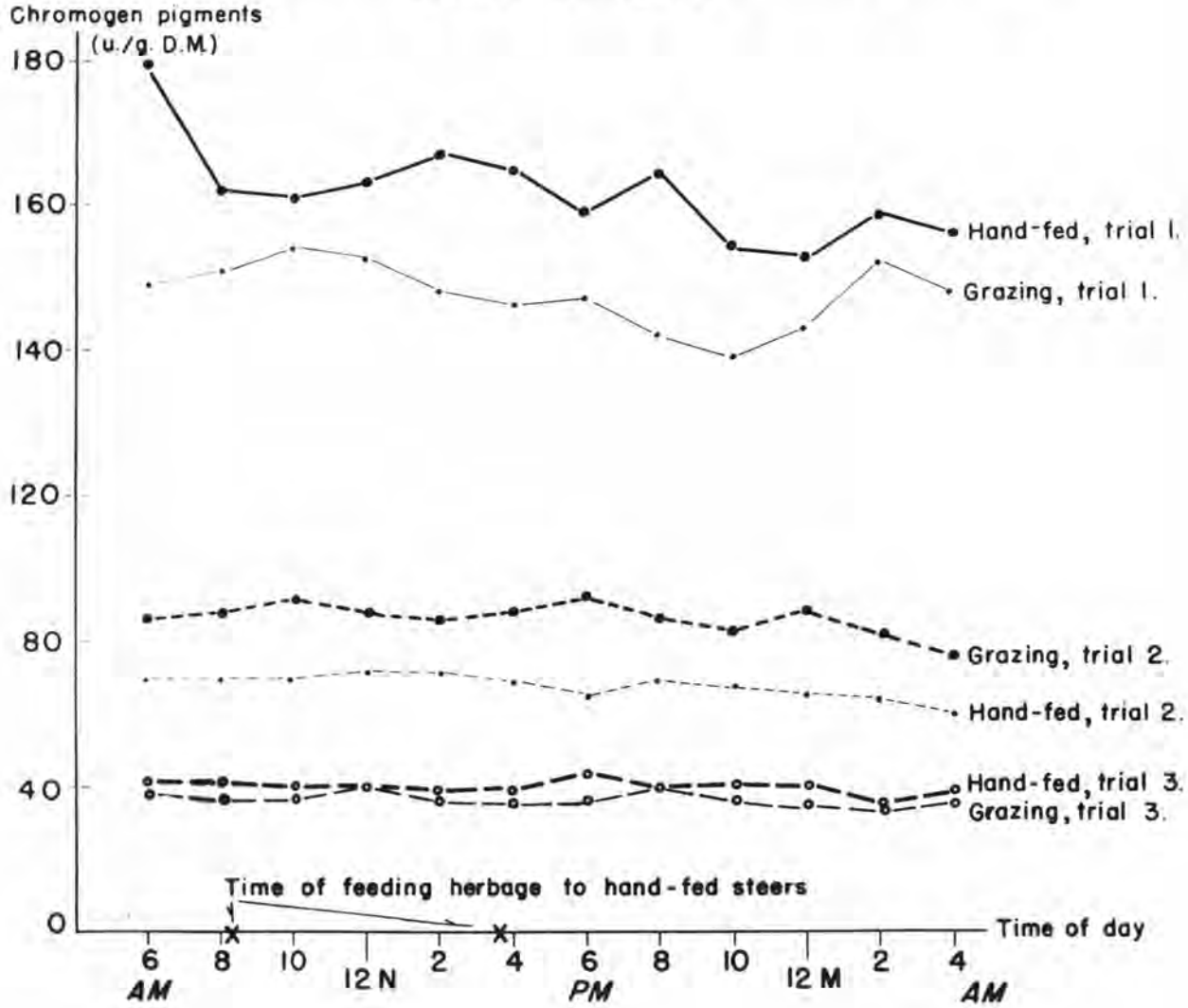
Figure 2 depicts the mean fecal chromogen concentration for the grazing and hand-fed steers at 2-hour intervals for each trial. No consistent excretion pattern of these pigments is evident with steers on either regime or in any of the trials. The mean fecal concentration in units chromogen per gram dry feces was 148 and 162 for grazing and hand-fed steers, respectively, for trial 1, 86 and 68, respectively, for trial 2, and 37 and 40 for trial 3.

Table 11. Average percentage recovery of chromogen pigments from hand-fed steers.

Trial No.	Steer No.	Total units[1] on dry basis consumed	excreted	Percentage recovery
1	2	2,412,432	1,701,272	70.52
	4	2,514,633	1,816,414	72.23
	6	2,350,229	1,692,043	71.99
	Mean			71.58
2	2	1,593,693	1,095,202	68.72
	4	1,551,118	1,047,009	67.50
	6	1,410,111	941,569	66.77
	Mean			67.66
3	2	725,291	698,179	96.26
	4	748,932	651,828	87.03
	6	670,110	729,508	108.86
	Mean			97.38

[1] Units are as defined by Reid *et al.* (54, p. 255-270).

Figure 2. Average Chromogen Excretion of Steers Grazing or Hand-Fed Crested Wheatgrass



The dry matter digestibility coefficients obtained by: (1) conventional consumption-excretion method, (2) chromogen ratio method, (3) fecal chromogen formula of Reid et al. (54, p. 255-270) and (4) forage chromogen equation of Petersen et al. (48, p. LVII) are presented in Tables 12, 13 and 14 for trials 1, 2 and 3, respectively. In addition, the digestibility coefficients determined by chromogen concentrations of feces collected at 6 a.m. and 4 p.m. and utilizing the Reid formula are presented for the 3 steers for the three trials in Table 15.

The average dry matter digestibility coefficients calculated by the chromogen consumption-excretion method were 78, 72 and 100 per cent of those obtained conventionally for trials 1, 2 and 3, respectively. Those determined by the fecal chromogen method were 79, 130 and 271 per cent of coefficients obtained in the usual manner for the three trials, and 92, 88 and 52, respectively when determined by the equation of Petersen et al. (48, p. LVII). Average digestibility estimates using bulked fecal samples taken at 6 a.m. and 4 p.m. were 79, 134 and 259 per cent of those coefficients obtained conventionally for trials 1, 2 and 3, respectively.

Dry Matter Digestibility Estimated by Fecal Nitrogen

The taking of "grab" fecal samples every two hours also allowed the examination of the intra-day nitrogen

Table 12. Per cent dry matter digestibility of immature Agropyron desertorum as determined conventionally and estimated by chromogen pigments.

Steer No.	Dry Matter Digestibility					
	Conventional	Consumption Excretion[1]		Fecal Chromogen Formula[2]		Petersen et al., Formula[3]	
	%	%	% of Conventional	%	% of Conventional	%	% of Conventional
1		52.3		50.8			
3		50.6		52.9			
5		50.9		52.9			
Mean		51.2		52.2			
2	67.0	51.8	77.3	52.9	79.0		
4	66.2	52.2	78.8	53.0	80.1		
6	68.4	53.0	77.5	53.0	77.5		
Mean	67.2	52.3	77.9	53.0	78.9	62.0	92.3

1 Formula chromogen = 80.158 units per gram dry matter by analysis. Chromogen concentration of forage of grazing steers assumed to be the same as that of hand-fed steers. Fecal chromogen concentration taken from composite fecal samples (obtained from total collections).

2 Forage chromogen calculated from the equation of Reid et al., viz.

Forage chromogen = 0.0925 fecal chromogen + 137.3383 log fecal chromogen - 242.1181

Fecal chromogen from composites.

3 Dry matter digestibility % = $96.6491 - \frac{310.0510}{\sqrt{\text{Forage chromogen}}}$

(units / gram dry matter)

Forage chromogen as given in footnote 1.

Table 13. Per cent dry matter digestibility of mature Agropyron desertorum as determined conventionally and estimated by chromogen pigments.

Steer No.	Dry Matter Digestibility						
	Conventional	Consumption Excretion[1]		Fecal Chromogen Formula[2]		Petersen' et al., Formula[3]	
	%	%	% of Conventional	%	% of Conventional	%	% of Conventional
1		46.0		64.2			
3		48.2		62.5			
5		49.2		61.9			
Mean		47.8		62.9			
2	58.0	59.6	102.8	67.1	115.7		
4	55.0	25.7	46.7	85.4	155.3		
6	58.8	38.9	66.2	70.6	120.1		
Mean	57.3	41.4	72.2	74.4	129.8	50.7	88.5

1 Forage chromogen = 45.486 units per gram dry matter by analysis. Chromogen concentration of forage of grazing steers assumed to be the same as that of hand-fed steers. Fecal chromogen concentration taken from composite fecal samples (obtained from total collections).

2 Forage chromogen calculated from the equation of Reid et al., viz.
Forage chromogen = 0.0925 fecal chromogen + 137.3383 log fecal chromogen - 242.1181
Fecal chromogen from composites.

3 Dry matter digestibility % = $96.6491 - \frac{310.0510}{\sqrt{\text{Forage chromogen (units / gram dry matter)}}}$
Forage chromogen as given in footnote 1.

Table 14. Per cent dry matter digestibility of dry-mature Agropyron desertorum as determined conventionally and estimated by chromogen pigments.

Steer No.	Conventional	Dry Matter Digestibility: Consumption Excretion[1]		Fecal Chromogen Formula[2]		Petersen' et al., Formula[3]	
	%	%	% of Conventional	%	% of Conventional	%	% of Conventional
1		52.9		135.9			
3		56.4		122.2			
5		40.0		191.7			
Mean		49.8		149.9			
2	51.2	50.4	98.4	145.8	284.8		
4	51.4	44.9	87.4	169.3	329.4		
6	53.5	61.1	114.2	105.7	197.6		
Mean	52.1	52.1	100.0	140.3	270.6	27.0	52

1 Forage chromogen = 19.849 units per gram dry matter by analysis. Chromogen concentration of forage of grazing steers assumed to be the same as that of hand-fed steers. Fecal chromogen concentration taken from composite fecal samples (obtained from total collections).

2 Forage chromogen calculated from the equation of Reid et al., viz.

$$\text{Forage chromogen} = 0.0925 \text{ fecal chromogen} + 137.3383 \log. \text{fecal chromogen} - 242.1181$$

Fecal chromogen from composites.

3 $$\text{Dry matter digestibility \%} = 96.6491 - \frac{310.0510}{\sqrt{\text{Forage chromogen}}}$$

(units / gram dry matter)

Forage chromogen as given in footnote 1.

Table 15. Per cent dry matter digestibility determined conventionally and as estimated by the formula of Reid et al. (54, p. 255-270), using bulked fecal samples collected at 6 a.m. and 4 p.m.

Trial No.	Steer No.	Dry Matter Digestibility		
		Conventional %	Fecal chromogen %	% of Conventional
1	1		52.9	
	3		52.9	
	5		53.4	
	Mean		53.0	
	2	67.0	53.0	79.1
	4	66.2	52.9	79.9
	6	68.4	53.0	77.5
	Mean	67.2	53.0	78.9
2	1		78.2	
	3		65.3	
	5		58.8	
	Mean		67.4	
	2	58.0	85.6	147.6
	4	55.0	67.6	122.9
	6	58.8	76.7	130.4
	Mean	57.3	76.6	133.7
3	1		171.7	
	3		154.0	
	5		157.8	
	Mean		161.2	
	2	51.2	142.0	277.3
	4	51.4	161.3	313.8
	6	53.5	102.2	191.0
	Mean	52.1	135.1	259.4

excretion in the feces of steers on both regimes. These results are graphically presented in Figure 3. No consistent diurnal excretion pattern occurred in fecal N excretion. The reason for the rapid increase in fecal N concentrations from 10 p.m. to 4 a.m. with the grazing steers during trial 1 and from 2 a.m. to 4 a.m. with the same steers in the second trial is not known definitely, but one might speculate that this is indicative of consumption of forage containing greater crude protein earlier in the day. The grazing steers excreted consistently greater amounts of N in trials 1 and 2 compared to their hand-fed counterparts suggesting efficient selection by these animals of higher protein herbage. In trial 3, where the herbage was dry and mature, the average fecal N excretion of the grazing steers was in the realm of that of the confined steers denoting limited opportunity for selective grazing.

Utilizing the method of Lancaster (36, p. 31-38), wherein digestibility was computed from fecal N concentration, yielded the dry matter digestibility coefficients presented in Table 16. Also presented in this table are the conventionally-obtained coefficients with the confimed steers for comparative purposes. Average dry matter digestibility in per cent, determined by the N-dry matter ratio and conventionally, was 65 and 67, respectively for trial 1,

Figure 3. Average Nitrogen Excretion of Steers Grazing or Hand-Fed Crested Wheatgrass

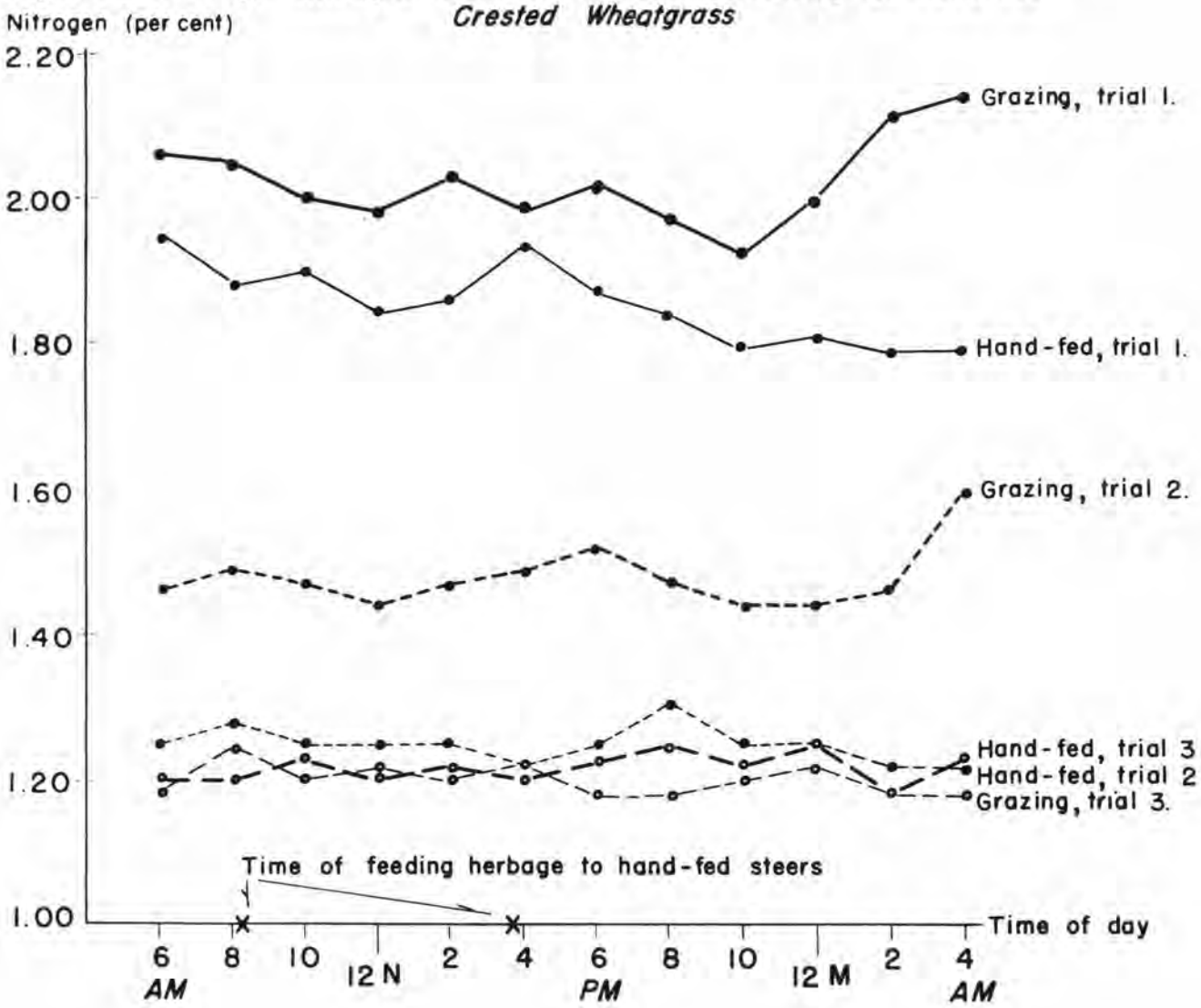


Table 16. Per cent dry matter digestibility determined conventionally and as estimated by the nitrogen-dry matter ratio method of Lancaster (36, p. 31-38).

Trial No.	Steer No.	Dry Matter Digestibility		
		Conventional %	N:Dry Matter Ratio[1] %	% of Conventional
1	1		69.7	
	3		67.6	
	5		69.6	
	Mean		69.0	
	2	67.0	65.7	98.0
	4	66.2	65.0	98.2
	6	68.4	65.4	95.6
	Mean	67.2	65.4	97.3
2	1		61.2	
	3		60.2	
	5		63.1	
	Mean		61.5	
	2	58.0	57.9	99.8
	4	55.0	53.0	96.4
	6	58.8	57.2	97.3
	Mean	57.3	56.0	97.7
3	1		47.2	
	3		46.6	
	5		52.7	
	Mean		48.8	
	2	51.2	52.5	102.5
	4	51.4	52.5	102.1
	6	53.5	55.2	103.2
	Mean	52.1	53.4	102.5

Table 16, continued

[1] Dry matter digestibility = $\frac{100\ n - x}{n}$ where n = per cent N in fecal dry matter and x = g fecal N/100 g. DM. intake. x = 60.03 for trial 1, 54.39 for trial 2, and 56.99 for trial 3. Individual data used for hand-fed steers.

56 and 57, respectively, for trial 2, and 53 and 52 for trial 3.

The estimation of dry matter consumption of grazing steers is dependent on the accuracies in the estimates of dry matter digestibility and fecal output; hence the errors in estimates of these measurements are compounded as errors in dry matter intake estimates. It would seem superfluous then, to tabulate consumption data utilizing all the various comparisons of fecal output estimates and the different comparisons of digestibility. Using all comparisons cited previously would result in 24 different values for dry matter intake. The data on fecal output estimates and digestibility estimates determined by the chromogen technique are erratic enough that any optimum procedure compatible to both for predicting intakes is not practical.

NON LIQUET
(Discussion)

Evaluation of Chromic Oxide

The results presented in the preceding section indicate certain problem areas that can be experienced using chromic oxide as a means of estimating the fecal production of range animals.

Failure to achieve recoveries of 100 per cent, at first glance, would indicate a limitation of this indicator. De facto, the emphasis on this area should be one of uniformity of recoveries. If percentage recovery were consistent both among animals and trial conditions, it would matter not that it was above or below complete recovery, since appropriate corrections could be applied. In the trials reported it is indicated that no justifiable correction factor is appropriate because of wide variations noted. That there may be a more rapid movement of Cr_2O_3 through the alimentary tract when the indicator is associated with higher digestible plant portions as proposed by Moore (45, p. 273-288) and others, might explain, in part, an over-recovery of Cr_2O_3. The results of trial 3, however, indicate that this is probably not the case since percentage recoveries of this trial were higher than those of the two trials where more digestible herbage was provided.

When the indicator was administered to stall-fed steers in cottonseed meal and as a Cr_2O_3-cellulose mix, the latter theoretically allowing a more uniform excretion than the free salt, recoveries were consistently below complete recovery. Fecal loss in total collections could account, in part, for the low recoveries in trial 5, since a new type of collection bag was being investigated. In trial 6, however, this was not the case for the new bags were abandoned in favor of larger and more firmly attached collection bags. Factors that may affect the diurnal excretion of the indicator as postulated by others, vide, time and mode of administration, frequency of dosing, and time of feeding or main periods of grazing, to name a few, could affect the percentage recovery.

The circadial variation in Cr_2O_3 excretion reported by others was substantiated with range animals. Such variation is also one of concern, but this is minimal if excretion patterns are consistently repeatable. This has not been the situation in results reported by others and was not observed in this study. It is apparent, then, that arbitrary fecal sampling times would not be possible under range grazing trials under the conditions as outlined in the first 3 trials. This shall be discussed at a greater length later.

No attempt was made in these trials to ascertain the

effect of certain factors on Cr_2O_3 excretion patterns. Hardison and Reid (26, p. 47) recognized some of these factors when they stated:

> "It seems possible that the excretion of Cr_2O_3 may be influenced by such factors as the degree of uniformity and time of forage intake, the specific gravity of Cr_2O_3, time of administering Cr_2O_3, the manner in which Cr_2O_3 is administered, plant species available for grazing, the stage of growth of herbage and such items as climatic conditions and management practices which may affect the grazing habits of animals. Also, the effects of water consumption, factors affecting the motility of the gastrointestinal tract, and the sex of the grazing animal upon the pattern of Cr_2O_3 excretion have not been examined."

Since that time many investigators have reported results of efforts to elucidate the significance of some of the above factors. Most of these studies, however, have been concentrated in areas of lush irrigated pastures and using grazing animals or confined animals fed forages derived from such areas. It is conceivable that many of the factors listed by Hardison and Reid could exert a greater influence on range trials where less rigorous controls may be exerted. Herbage amounts, quality and species, for example, would be more variable under dry-land conditions than under irrigated or higher rainfall conditions.

Trials 5 and 6 took advantage of more recent published data that, in general, indicated a greater success by increasing the number of doses of Cr_2O_3 per day and

employing different means of providing the indicator. These two trials were initiated to further evaluate the efficacy of the indicator based on the premise that a Cr_2O_3-cellulose mix might be more uniformly mixed with rumen contents, thus allowing a less variable excretion of the indicator. The fecal sampling procedure, as outlined in a previous section, was patterned from results of others, wherein certain success was achieved in estimating the fecal production of animals, using fairly set "grab" sampling times. Since there have been differences of opinion as to the most appropriate times, and, since there have been no reports to indicate when fecal samples should be obtained for best results in studying forage indigenous to range areas, sampling times were set that were compatible to prevailing husbandry practices. Under stall-feeding conditions and using a feed indigenous to the range area, results indicated that fecal output estimates were in error from actual fecal production by 0.7 to 0.8 pounds per day. If actual dry matter intake were 10 pounds daily and there was no error in estimate of digestibility estimates, this error would be 7 to 8 per cent in any estimates of dry matter intake.

The procedures used in trial 6 were employed in a cross-over trial on range herbage, Agropyron desertorum. The indicator was supplied in a cellulose mix to steers

twice daily and fecal sampling times were the same as in trial 6. Since the author was not directly responsible for the conduct of the trial and the collection of the data, such information is not presented in the body of this dissertation. The data are, however, summarized in the addendum. This summary indicates that estimates of fecal production of grazing steers were in good agreement with measured fecal outputs - 0.06 pounds per day when steers were supplemented with barley and 0.12 pounds per day when steers received no supplement. In addition, the per cent recovery of Cr_2O_3 was quite complete, i.e., 99.6 per cent from supplemented steers and 102.2 per cent from those not supplemented.

Results of the recent grazing trial are the most encouraging so far. They suggest that the indicator has more potential in range intake studies than has been demonstrated heretofore by the application of experimental procedures that have been acceptable in other areas. However, the Cr_2O_3-cellulose mix has not been investigated in other sections of the range region, and, though it was practical under one set of conditions, it may not be under others.

The protean results obtained with Cr_2O_3 in these trials, as well as in other reports in the literature, include erratic results in: (1) percentage recovery,

(2) non-repeatable circadial excretion, and (3) the estimates of fecal production. In addition, recognition must be given to the factors that could affect the potential of the material as stipulated by Hardison and Reid (26, p. 35-52). Such vacillating results would seem to reaffirm the statement of Balch et al. (1, p. 196) viz:

> "..... It is therefore recommended that the suitability of the sampling times should be checked against complete collection of faeces under the conditions of each experiment. This would seem to be especially necessary where unusual crops or unusual grazing management are being used."

Evaluation of Apparent Dry Matter Digestibility Using Chromogen Pigments and Fecal Nitrogen

Since the original reports of Reid et al. (53, p. 60-71; 54, p. 258-270), a wealth of information has appeared in the scientific literature relative to the use of plant chromogens to estimate digestibility of pasture herbage. As stated earlier the use of chromogen pigments has in some cases been an acceptable method of estimating herbage digestibility. However, in the hands of other investigators this indicator has proven ineffective. Evidence has appeared in the literature to reduce the application of this particular indicator. Results reported by Irwin et al. (30, p. 541-551), Davidson (16, p. 5; 17, p. 86-92) and Mixner et al. (44, p. 67-74) indicate that these pigments are digestible, a fact that greatly affects their

usefulness in assessing herbage digestibility. Analytical difficulties have been stressed by Lancaster and Bartrum (38, p. 489-496) and others. Such difficulties in analysis minimize the effectiveness of the indicator and thus it does not meet the conditions of an indicator as set forth by Raymond and Minson (52, p. 283) when they stated that an indicator should be ".... readily analyzed by physical or chemical methods, ...". These analytical problems may also indicate that a high degree of competence attained by extensive experimentation in this field is necessary to obtain satisfactory results using this indicator.

Much of the discourse on chromic oxide is applicable to a discussion of the internal indicator. The erratic results achieved using chromogens in the first three trials indicate the limitations of such a procedure in range grazing trials.

Although there was no consistent excretion pattern exhibited among steers or trials, variation in concentrations were great enough that repeatable digestibilities would be difficult to obtain. It would seem that any time for taking "grab" samples would be adequate under these conditions to arrive at "some means of a digestibility estimate". Such an assumption is dangerous and inadvisable where precision and accuracy are requisites.

Dry matter digestibility estimates using chromogens

were uncompromisingly in error as compared to those obtained conventionally with hand-fed steers. None of the procedures of calculating digestibility resulted in adequate estimates except that proposed by Petersen and co-workers of the Oregon Agricultural Experiment Station. This equation failed, however, to provide comparable estimates in trial 3. Since Reid and co-workers established their equation on forages containing 100 units chromogen per gram dry matter, and since none of the forages in this study reached that amount even in June, it is probable that such an equation is not applicable under these conditions. This was emphasized by Petersen et al. (48, p. LVII), when they reported that a graphical application of data resulted in the same digestibility being predicted for forages of different chromogen content with the regression line passing through a minimum of 80 units of chromogen per gram of dry matter. The data presented herein emphasize the ineffectiveness of this indicator when forages contain less than 100 units per gram on a dry basis. Apparently the only time that range herbage would be above this value would be in early spring.

The basic disagreement in using the equation set forth by the Oregon workers is the problem in sampling herbage that is representative of that consumed by grazing animals. There have been proposals offered to circumvent

the difficulties in selective grazing. Weir and co-workers (59, p. 235-237) presented a review on the use of the esophageal fistula technique for studying selective grazing and digestibility of herbage by sheep and cattle. Lesperance et al. (40, p. 682-689) discussed the use of both the esophageal and rumen fistulae as means of evaluating grazed forage. Workers at the Oregon Experiment Station have studied the effectiveness of rumen clearance as a means of assessing the quality of range herbage, specifically, Agropyron desertorum. Employing these procedures and the Petersen equation could conceivably result in comparatively accurate estimates of dry matter digestibility.

The use of the fecal nitrogen:dry matter ratio as a "tool" for estimating dry matter digestibility seems to offer more promise than "chromogen" pigments. Dry matter digestibility estimates varied from 1 to 4 per cent among trials from those calculated from the conventional trial conducted simultaneously with steers hand-fed crested wheatgrass. This difference is not of great importance when one considers the variation of forage quality among the trials. Although the relationship between fecal nitrogen and dry matter digestibility has been established, as mentioned previously, and this procedure has produced "reasonable" results in range trials, the method

also has limitations. Some of these limits have been stated before and briefly these may be summarized by stating that the fecal nitrogen:dry matter ratio varies as to different species of animals and different types of feed. The establishment of regression equations of fecal nitrogen on dry matter digestibility might offer some means of circumventing the basic confines, but as Harris et al. (27, p. 233) emphasized:

> "If chromogens or nitrogen are relied upon to determine intake and digestibility each investigator should determine the appropriate regression equations to use under his own conditions."

To be of most reliable value such regression equations for range conditions should encompass a variation in forage quality with data collected over a number of years.

Whatever method is utilized, the paramount criterion is that it provides repeatable and reliable estimates of digestibility.

As in the case of the excretion of chromogenic substances there was no consistent pattern of excretion of nitrogen either among steers on either management scheme, or among trials. Reference to Figure 3 indicates that, except for the early morning hours of 2 and 4 a.m., "grab" fecal samples may be taken at any time if fecal nitrogen is to be used as an indicator of dry matter digestibility.

Fecal nitrogen concentration was higher for steers during the first trial and this might be explained by the fact that herbage was higher in nitrogen than that of the later trials. Selective consumption by the grazing steers in trial 2 may explain why fecal nitrogen concentrations of these steers were higher than those of their hand-fed counterparts. The restriction of the grazing steers to small areas, however, would seem to preclude much selective grazing. The disagreement between the fecal nitrogen concentration of the steers on the two regimes in trial 2 is indicative of a problem that exists in trying to duplicate the consumption of grazing steers by clipping and feeding "similar" herbage to confined steers.

The presentation in this section has been directed to a discussion of the more succinct and pertinent areas exemplified by the data. As in other research endeavors further extension and interpretation of the data are possible, but such expansion would not increase the accuracy for which the indicators are to be used. Further statistical development of the data, e.g. correlations or regression equations, is not justified unless this was an established objective prior to initiation of the trials.

FINIS CORONAT OPUS
(Summary)

1. The external indicator, chromic oxide, was investigated as to its reliability in estimating the fecal production of steers consuming forage indigenous to the sagebrush-bunchgrass region. The results of 5 trials are reported. Three trials were conducted on improved range consisting predominately of crested wheatgrass Agropyron desertorum. The other 2 were conducted with rush-sedge hay.

2. One criterion employed in the evaluation of an indicator is its relative recovery in the feces of test animals. The percentage recovery of chromic oxide ranged from 94 to 126 in the first trial and 99 to 114 and 112 to 138 in trials 2 and 3, respectively. Recovery was not consistent among animals in succeeding trials. In trials 1 and 2 the mean recovery percentage for the hand-fed animals was 102 and 101, respectively. These values were the closest to 100 per cent of the 3 trials.

3. Excretion patterns of chromic oxide were established for the 6 steers in the first 3 trials by collecting "grab" fecal samples every 2 hours for the 5 day collection period. The concentration of the indicator exhibited a definite circadial pattern but this pattern

varied between animals on either regime, i.e. hand-fed or grazing wheatgrass, within any one trial. There were also differences in the excretion pattern among the various trials. The nonrepeatability of such excretion patterns under the conditions outlined in preceding sections, within one season, indicates that there is no consistently reliable time for fecal sampling. In addition, the establishment of regression equations for one set of environmental conditions may not be applicable under another set.

4. The estimates of fecal production using chromic oxide, as compared to that measured by collection bags was variable among the steers and the 3 trials on crested wheatgrass. Estimates varied from 20 per cent underestimation to 49 per cent overestimation in trial 1, from -22 to +47 per cent and from -34 to +54 per cent in trials 2 and 3, respectively. Also reported are the results of "bulking" "grab" fecal samples collected at 6 a.m. and 4 p.m. and the fecal production as estimated by the mean concentrations of all "grab" samples. Estimates using these latter two methods deviated more from measured fecal output than the estimates between the 3 grazing steers and those hand-fed using the 3 estimating procedures.

5. The latter 3 trials utilized information that had appeared in the literature after the first 3 trials were conducted. This information indicated, in general, better results with chromic oxide if the indicator were given more than once daily. In addition, other means of administering the material, especially using certain carriers, seemed to offer much promise. Trial 5 was initiated to examine the efficacy of 3 different means of administering the indicator in estimating the fecal output of steers fed rush-sedge hay. Supplying chromic oxide in cottonseed meal, "free" material in capsules, and as a chromic oxide - cellulose mix resulted in estimates of fecal production of 119, 138 and 123 per cent of measured outputs, respectively. Average per cent recovery for the 3 groups was 84, 73, and 81 for cottonseed meal as a carrier, "free" chromic oxide and chromic oxide - cellulose, respectively. Trial 6 was initiated to further evaluate the chromic oxide - cellulose mix as influenced by the quantity of hay consumed. Fecal production was estimated within an average of 0.7 - 0.8 lb. per day. Recovery of the indicator in this trial averaged about 85 per cent.
6. A trial was conducted on crested wheatgrass range using the procedures as outlined for trial 6. Results

of this trial are the most encouraging and applicable to date. Fecal production was estimated within about 0.1 lb. per day of measured values. Recovery of the indicator was essentially 100 per cent. This last trial suggests that a chromic oxide - cellulose mix may be the most applicable method of using the external indicator in range nutrition studies. However, investigations need to be continued in order to support or refute this supposition.

7. The possibility of using the chromogen pigments as a means of estimating dry matter digestibility of range forage was also investigated in trials 1, 2, and 3. Recoveries of these pigments from hand-fed steers were highly variable ranging from 67 to 109 per cent among trials with mean recoveries of 72, 68, and 97 per cent for trials 1, 2, and 3, respectively. "Grab" sampling as mentioned above showed that there was no apparent diurnal excretion of the pigments with steers on either regime or in any of the trials. Dry matter digestibility coefficients using these pigments were computed in different ways and compared to those obtained conventionally with the hand-fed steers. Coefficients were so variable as to be almost meaningless. In trial 3 coefficients obtained by the chromogen

consumption-excretion method agreed precisely with those obtained by conventional means. The application of an equation for obtaining dry matter digestibility using forage chromogen content was presented and its possible application to range livestock nutrition studies were discussed. Because of the vascillating results obtained in the 3 trials using plant chromogens it would appear that they are of negligible value for range investigations.

8. The applicability of the nitrogen:dry matter ratio as a means of estimating dry matter digestibility of steers consuming range forage was also studied in trials 1, 2 and 3. This procedure resulted in digestibility estimates that agreed very favorably to those determined conventionally with the hand-fed steers. However, the application of the constants determined with the confined steers to fecal nitrogen values obtained from grazing steers resulted in variable estimates. This would generally be the expected result when one attempts to apply certain constants obtained with one group of animals to another group.
9. The variable results reported for the estimation of fecal production and estimates of dry-matter digestibility preclude the value of determining the dry matter

consumption of steers on range forage since any intake figures are dependent upon the accuracy of each of the estimates and are influenced by the type of fecal sampling procedure employed.

10. More basic information on the quality and quantity of range forage is required. Indicators, both external and internal, in combination, can be valuable "tools" in this field of research. The improvement of procedures in supplying chromic oxide seems to increase the potential of using this material in range nutrition studies. The use of fecal nitrogen as an internal indicator to estimate dry matter digestibility of range forage species also appears promising. Application of more recent techniques in ruminant nutrition studies, i.e. sampling via esophageal and rumen fistulas can also contribute to progress in this field.

ADDENDUM

A cross-over trial, utilizing steers grazing Agropyron desertorum, was conducted to ascertain the effectiveness of Cr_2O_3 in estimating fecal production when steers were supplemented with barley. As outlined previously, the indicator was supplied in a cellulose mix twice daily and fecal sampling times were the same as in trial 6.

The results of the grazing trial are presented in Table 17. Barley supplemented at 2 lb. daily did not alter the accuracy of estimated fecal output compared to that obtained by total collections. Mean fecal dry matter output was estimated within 0.12 and 0.06 lb. per day with steers unsupplemented and supplemented with barley, respectively. The average recovery of Cr_2O_3 was 102.2 per cent for the unsupplemented steers and 99.6 per cent for those supplemented.

Table 17. Effects of barley supplementation on estimates of dry matter fecal output (D.M.F.O.) by Cr_2O_3 in a cellulose carrier with steers grazing Agropyron desertorum.

Treatment	Mean Measured D.M.F.O. (lb./day)	Mean Estimated D.M.F.O. (lb./day)	Recovery Cr_2O_3 (%)
No Barley	8.46	8.34	102.2
Barley	9.23**	9.29*	99.6
LSD 5%	0.46	0.89	7.8
1%	0.62	1.20	10.5

* Significantly different ($P < 0.05$) from estimated output of steers not supplemented.

** Significantly different ($P < 0.01$) from measured output of steers not supplemented.

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