SOME ASPECTS OF MINERAL NUTRITION IN RELATION TO BISON FLAX

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(WITH THREE FIGURES)

Introduction

That the yield and quality of oil produced by flax seed varies from year to year is a matter of common knowledge. In the absence of exact experimental data it is frequently stated that climatic factors are responsible for this variation. Since little is known about the actual process of oil synthesis by the flax plant and still less about the effect of specific environmental factors on this synthesis, it was felt that some careful studies under controlled conditions should be initiated. In these first studies the authors undertook to grow seed flax in selected nutrient solutions in an attempt to determine if there was any relation between mineral nutrition and the quality and quantity of the oil produced from flaxseed.

Fertilizers and their relation to yield of flax grown in the field have received some attention by various workers throughout America; little exists in the literature, however, relative to the growth of flax in nutrient solutions. Turner (15) studied the effect of variations in the nitrogen supply on the ratio of top to root growth of Winona flax. Using solutions high and low in nitrate nitrogen, he found that there was little difference in the nitrogen content of flax grown in these nutrients. He attributed the results obtained to relatively limited use of nitrogen by flax, and concluded that in many cases nitrogen fertilizers when applied to flax are not particularly beneficial.

After studying the effect of the light factor on cell production of fiber flax grown in sand culture, Robinson (8) reported that the tallest growth resulted when fiber flax was grown in ten hours of light per day; flax which was exposed to light eighteen hours each day matured earliest but produced the least vegetative growth. Thus the growth period of flax might be shortened by lengthening the photoperiod. His report showed that for fiber flax nitrogen was particularly important in early growth, and that potassium was more important as the plants matured. Adams (1) worked out the relation between duration of light and growth, with reference to flax grown in soil, and his findings in most instances bear out those of Robinson.

In Germany, Selle (11) made tests on soil acidity in relation to growth of fiber flax. He noted that the yields, based on fiber, were greatest on lime-rich soils and poorest on acid soils. Potassium, according to Powers (7), was of the greatest importance in fertilizers for fiber flax; its presence...
in the soil increased strength and length of fiber and vigor of growth. Nitrogen was second in importance. Potash was required relatively early in order to obtain maximum benefits.

Fiber flax was grown in liquid nutrient media by Shkol’nik (12). He worked especially with boron, manganese, aluminum, and copper. Boron was found to be of paramount importance, its absence causing a poor development of the root system and eventual death. The optimum concentration of boron was 0.5 mg. per liter; doses of about 5 mg. per liter were toxic. Manganese and other elements mentioned appeared less important. A few years ago Sommer (13) showed that the addition of small quantities of copper to nutrients supporting flax growth resulted in increased vigor as compared to growth of plants in solutions lacking that element.

Schmalfuß (9) grew fiber flax in pots of loamy soil, the water content of which was controlled, and which received various fertilizer treatments. He reported the following findings: lignification of fiber cells was increased by lowering the water content of the soil, and also by increasing the amount of sulphate ion added; lignification was decreased by increasing the amount of chloride ion; the fiber content of the stem was increased by nitrogen, and more so by K₂O; the fiber content was increased by the sulphate ion, and decreased by the chloride ion. He further reported that the yield and quality of oil produced were affected by fertilization. In this respect the iodine number of oil from nitrogen-starved plants was highest. The iodine number was affected more by anions than by cations; the chloride ion increased and the sulphate ion decreased the iodine number of the oil produced. His results showed that the nitrogen content of the seed varied inversely with the oil content. The observed reactions on amount and quality of fiber and oil he explained as results of effects of the above mentioned factors on the colloids, hydration of the plant cells, and the water economy of the plants.

No attempt will be made here to present an extended review of the literature on the subject of physiological nutrient solutions or on the technique of nutrient culture work. Tottingham (14) gave a thorough review of the subject in his paper of 1914, and since then a number of workers have dealt with various other phases. The triangle system as used in this work has been adequately discussed by Schreiner and Skinner (10) and Miller (6).

Preliminary experiments were carried out in the winter of 1935–36 in order to find the approximate concentration of nutrients that would support the growth of flax. In these tests a pure line of Bison flax, developed by H. L. Bolley and O. Heggeness of the North Dakota Agricultural Experiment Station was grown to the flowering stage in Shive’s type I, three-salt, liquid nutrient solutions. The best growth was obtained in solutions
having a composition of 25 to 37.5 per cent. \( \text{KH}_2\text{PO}_4 \), 50 to 62.5 per cent. \( \text{Ca(NO}_3\text{)}_2 \), and 25 to 37.5 per cent. \( \text{MgSO}_4 \), based on the total salt concentration of the nutrient solution.

**Methods and materials**

During the winter of 1936–37 Bison flax was grown to maturity in ten of Shive’s type I, three-salt liquid nutrient solutions. These solutions were chosen so as to be well distributed over the triangle; they included solutions R1S5, R2S1, R2S3, R2S4, R2S5, R3S2, R3S3, R4S2, R4S3, and R5S2. Partial volume molecular concentrations are given in table II.

The necessary elements for plant growth are supplied in type I, three-salt solutions by \( \text{KH}_2\text{PO}_4 \), \( \text{Ca(NO}_3\text{)}_2 \), and \( \text{MgSO}_4 \). In making up the nutrients these salts were added from M/2 stock solutions to distilled water in the proportions prescribed by the committee of Biology and Agriculture of the National Research Council in its 1919 report. The osmotic values of all solutions were determined cryoscopically and were found to be somewhat under one atmosphere.

Equal amounts of iron, manganese, and boron were added to each nutrient solution. The iron was added as soluble ferric phosphate from a 0.0134 M stock solution, 2 ml. of this solution being added to each liter of nutrient. One-half part per million of \( \text{MnSO}_4 \) and \( \text{H}_3\text{BO}_3 \) was added to each liter of nutrient solution prepared. All chemicals used in preparing the nutrient solutions were of C. P. analyzed grade secured from the J. T. Baker Chemical Company. Fresh solutions were prepared and added to the cultures daily.

A constant renewal system (fig. 1) was set up and regulated to feed, at the rate of 1.5 liters of solution per day, into each series of cultures. In this system the fresh nutrient was added to the upper reservoir, and from there it passed through a constant-level siphon, into the lower reservoir. The nutrient then flowed through the delivery siphon, dropped into the intake tube, and flowed down the tube into culture A. When the level of solution in culture A exceeded that in the delivery arm of the siphon, the solution was siphoned from culture A into culture B; the same process occurred in cultures B and C until the nutrient solution passed from that set of cultures into the drain. Each drop of nutrient, upon falling into the delivery tubes, would trap an air bubble, which was driven down through the intake tube, aerating the nutrient solution as it rose to the surface in each culture jar. A shield of insulation board was constructed to cover the reservoirs in order to prevent heating of the solutions in the reservoirs by the sun’s rays. The term “bank” has been used to designate the three culture jars of a series. The two one-gallon reservoirs of each bank were closed with rubber stoppers and painted to exclude light, as was all glassware used in the set-up.
The paraffined stoppers, used to support the plants, were anchored in tin, two-quart Economy jar covers which were heavily coated with paraffin. Ten such banks, each composed of two reservoirs and three cultures, were used in the experiment. These banks remained in a fixed position during the entire study.

FIG. 1. Constant renewal system used in flax nutrition studies.

Seed of the Bison variety from the same lot used in the 1935–1936 studies was used in this experiment. Quantities of seedlings were obtained using methods similar to those described by Hoagland and Broyer (4). When the tops of the seedlings were 3 cm. long, uniform seedlings were selected, their stems were wrapped in cotton at the base and enclosed in a cork. Five seedlings were placed in each culture container. Cultures were set up on November 27, 1936. The plants were supported by a network of threads. The temperature of the greenhouse was thermostatically controlled at 18 to 20° C.; the relative humidity generally ranged from 40 to 50 per cent.

Throughout the experiment the plants were exposed to 24 hours of light each day. At night light was supplied by four 200-watt Mazda bulbs equipped with porcelain reflectors. As measured by the Weston Photronic cell a light intensity of from 130 to 140 foot-candles was obtained all along the table at the tops of cultures of each bank.

Hydrogen ion determinations were made on the freshly prepared nutrient solutions and the solutions leaving the different banks on three different days during the period of the experiment using both the Youden quinhydrone cell and colorimetric procedures. In all cases these tests were conducted according to the directions given by Clark (3).

All cultures were allowed to grow to maturity or until the main stems of the plants had completely dried. Upon removal of the plants from the
cultures, data on oven-dry weight of roots and tops were taken on the five plants from each culture. Dry weights were determined on material dried to constant weight at 70°C. After the seeds had been removed, fruits were hand picked and threshed, the seed from the main stems being kept separate from that of the lateral branches. A record was made of main stem, lateral, and total seed in each culture; and the weight of one hundred seed samples of the main stem seed was also recorded.

The flaxseed samples were stored together with a bulk sample and all assumed to acquire the same moisture content as the bulk sample. Moisture determinations were run on the bulk sample since the experimental seed lots were too small to permit of such determinations. Each entire experimental lot was then weighed and thoroughly ground with round sand in a porcelain mortar. The ground seed and sand was carefully washed with Skelly Solve F into a special glass extraction thimble and extracted for 24 hours in a percolation extractor with the same solvent. The quantity of oil was determined by direct weighing of the oil in the tared extraction flask after removal of the solvent and drying in a vacuum oven for one half hour at 110°C. The iodine number of the extracted oil was determined refractometrically according to the method of Hopper and Nesbitt (5).

Total nitrogen was determined on the extracted residue by the official A.O.A.C. method.

Results and discussion

The hydrogen ion concentrations of the nutrient solutions were determined on freshly prepared solutions on January 17, February 28, and March 26. All of the fresh nutrients had pH values between 5.7 and 6.0. Likewise, the solutions coming out of the different culture jars were tested

### TABLE I

<table>
<thead>
<tr>
<th>Solution and Date of Test</th>
<th>pH Value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh solution</td>
<td>R1S5</td>
</tr>
<tr>
<td>January 17</td>
<td>6.05</td>
</tr>
<tr>
<td>February 28</td>
<td>6.15</td>
</tr>
<tr>
<td>March 26</td>
<td>5.35</td>
</tr>
</tbody>
</table>

* Average of three cultures in bank.
on January 17, and February 28, fifty-one and seventy-five days, respectively, after the seedlings had been placed in the culture jars (table I). At the time the used solution was first tested, the plants were just entering the period in which they made most marked and rapid growth. The pH of solutions coming from the different cultures of a bank was uniform, and showed little change from that of the freshly prepared nutrients. The second test was made after the plants had attained a considerable size and were still growing rapidly. The results in the second test showed that the pH of the nutrients varied slightly as they left the different cultures of a bank; there was a general tendency, however, for the solutions to become slightly more acid as they flowed through the banks.

These changes seem to be in no way correlated with the original composition of the nutrient solutions. All solutions were uniform in that they tended to become more acid. CHIZHERSKAIA (2) carried on water culture experiments to determine the relation of sprout growth in flax to the pH of the nutrient medium. From these experiments she found that the maximum sprout growth occurred at pH 5, and pH 9. It must be remembered that her conclusions on acidity relations were based upon the early growth of flax, while in the experiments reported above large plants were involved.

Considering the nutrients on the basis of the length and dry weight of tops produced, it is evident that a slightly greater growth resulted in cultures containing relatively high concentrations of KH$_2$PO$_4$ (table II). In general there was no region of the triangle used where nutrients were definitely superior in respect to height and dry matter produced. There is no indication of a clear cut correlation between the composition of the nutrient solution and the final length of top growth; the cultures which were slowest to mature, however, R1S5 and R5S2, made the tallest top growth. Plants in all cultures were considerably taller than field grown plants of the same variety. According to the records available in this Experiment Station, this same strain, over a period of years, has produced plants ranging from 40 to 61 centimeters in height (data obtained from H. L. Bolley and O. Heggheeness). The data on dry-matter produced can be considered only as relative owing to the fact that dry-weights could not be taken until the plants had matured, with a consequent loss of some leaf material. Yields in regard to dry-weight were rather definitely related to the concentration of KH$_2$PO$_4$, the largest amount of dry-matter being obtained in solutions with the highest concentration of that salt. Concentration of other salts in these high KH$_2$PO$_4$ cultures varied considerably. The data appear to indicate that this variety of flax is not a heavy user of nitrogen.

The date of maturity of plants growing in the different solutions showed a certain degree of relationship to the amount of KH$_2$PO$_4$ present. Those solutions relatively high in KH$_2$PO$_4$ produced the earliest maturing plants.
<table>
<thead>
<tr>
<th>BANK NO.</th>
<th>SOL’N NO.</th>
<th>KH₂PO₄</th>
<th>Ca(NO₃)₂</th>
<th>MoSO₄</th>
<th>GROWTH PERIOD</th>
<th>HEIGHT OF TOP</th>
<th>DRY MATTER PER PLANT**</th>
<th>AVERAGE TOTAL SEED PER PLANT†</th>
<th>AVERAGE WEIGHT PER 100 MAIN-STEM SEEDS</th>
<th>PERCENTAGE OIL CONTENT</th>
<th>IODINE NUMBER</th>
<th>MEAL BASIS</th>
<th>OIL AND WATER FREE BASIS</th>
<th>PERCENTAGE CRUDE PROTEIN (N x 6.25)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>R185</td>
<td>0.0022</td>
<td>0.0108</td>
<td>0.0043</td>
<td>185</td>
<td>119.4</td>
<td>2.566</td>
<td>0.428</td>
<td>0.641</td>
<td>36.6</td>
<td>164</td>
<td>27.6</td>
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<td>2</td>
<td>R281</td>
<td>0.0053</td>
<td>0.0027</td>
<td>0.0132</td>
<td>169</td>
<td>106.7</td>
<td>2.267</td>
<td>0.519</td>
<td>0.633</td>
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<td>0.0047</td>
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<td>0.0071</td>
<td>159</td>
<td>109.2</td>
<td>2.378</td>
<td>0.611</td>
<td>0.663</td>
<td>37.2</td>
<td>174</td>
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<td>0.0090</td>
<td>0.0045</td>
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<td>R285§</td>
<td>0.0041</td>
<td>0.0104</td>
<td>0.0021</td>
<td>149</td>
<td>97.8</td>
<td>1.487</td>
<td>0.342</td>
<td>0.636</td>
<td>37.2</td>
<td>175</td>
<td>27.9</td>
<td>48.84</td>
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<td>0.0048</td>
<td>0.0072</td>
<td>149</td>
<td>111.8</td>
<td>2.249</td>
<td>0.695</td>
<td>0.672</td>
<td>37.7</td>
<td>181</td>
<td>24.8</td>
<td>43.86</td>
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<td>7</td>
<td>R383</td>
<td>0.0068</td>
<td>0.0068</td>
<td>0.0045</td>
<td>177</td>
<td>99.0</td>
<td>2.899</td>
<td>0.719</td>
<td>0.661</td>
<td>36.1</td>
<td>172</td>
<td>26.7</td>
<td>45.85</td>
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<td>8</td>
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<td>0.0094</td>
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<td>0.0047</td>
<td>177</td>
<td>109.2</td>
<td>3.037</td>
<td>0.687</td>
<td>0.686</td>
<td>36.6</td>
<td>175</td>
<td>26.9</td>
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<tr>
<td>9</td>
<td>R483</td>
<td>0.0090</td>
<td>0.0068</td>
<td>0.0022</td>
<td>174</td>
<td>109.2</td>
<td>2.927</td>
<td>0.655</td>
<td>0.660</td>
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<td>26.9</td>
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<td>10</td>
<td>R582</td>
<td>0.0118</td>
<td>0.0047</td>
<td>0.0023</td>
<td>183</td>
<td>120.7</td>
<td>3.248</td>
<td>0.672</td>
<td>0.662</td>
<td>36.1</td>
<td>174</td>
<td>27.1</td>
<td>46.58</td>
<td></td>
</tr>
</tbody>
</table>

* Average value per plant from 15 plants in all three cultures of each bank.

** Exclusive of chaff.

† Total lateral and main-stem seed.

‡ Moisture assumed at 5.68% on basis of other samples stored under same condition and analyzed.

§ Figured on basis of culture 1 only.
Plants in high-nitrogen cultures tended to be more vegetative. Cultures high in KH$_2$PO$_4$ and relatively high in nitrogen were intermediate as regards maturity. Magnesium sulphate seems to have played a less positive rôle as far as length of growth period is concerned. The character of growth is shown in figures 2 and 3.

**Fig. 2.** The appearance of the top growth in the cultures of banks 1, 2, and 3 on January 1, 1937.

From the standpoint of total weight of seed produced, solutions medium to high in KH$_2$PO$_4$ again showed the best yields (table II). The presence of medium to high concentrations of the nitrogen-carrying salt seemed to depress the total yield of seed unless the amount of acid phosphate present was also fairly large. Data obtained on the yield from plants growing in the field showed those plants had produced, over a period of years, about 1 gram of seed per plant. In this experiment the yields in the different cultures ranged from 0.342 to 0.719 gm. per plant which was considerably less than the yields obtained from field grown flax of this variety.

The weight of 100 average seeds produced on the main stems of the plants in the different nutrient media ranged between 0.633 and 0.686 gm. Seeds from a large number of samples of Bison flax that were grown in the
field in different localities varied from 0.318 to 0.677 gm. per 100 seeds and averaged about 0.533 gm. per 100 seeds. Our seed with an average of 0.657 gm. per 100 seeds was generally heavier than seed from this variety grown on plants in the field. The general tendency was toward heavier seed in cultures relatively high in KH₂PO₄; whereas plants grown on the nitrogen-high nutrient tended to produce lighter seed. All seed produced was, however, surprisingly uniform in weight (table II).
Analysis of the seed produced during this study showed that the yield of oil was about equal to that of the average field-grown seed, and that the iodine number of the oil was comparable to the highest of oil produced in field-grown flax at this station (table II). The amount of oil produced with the different nutrient solutions was rather constant, varying from 36.1 to 37.9 per cent., when calculated on a 5.68 per cent. moisture basis. Commercial samples of Bison varied from 32.0 to 40.0 per cent. with an average of 37.0 per cent. when calculated on the same basis. Tests for iodine number, made on the seed harvested from plants grown in this experiment, gave a range of from 164 to 181. Oil having the highest iodine number was produced in cultures well supplied with potassium acid phosphate. Data from the records on field tests conducted at Fargo show oils from flax-seed produced on Bison flax grown in the field had iodine numbers of 161 to 167. These findings indicate that the degree of saturation of linseed oil is not a fixed character but varies with the nutrient supply available.

The protein content of the seeds from the main stems of plants ran uniformly high in all cases, ranging from 38.36 to 49.63 per cent, on an oil and water free basis. Other experimental samples from field grown material have been found to vary from 28.4 to 53.4 per cent, crude protein on the same basis. Cultures high in Ca(NO₃)₂ and fairly high in KH₂PO₄ produced the highest yields of protein although the total difference was not great. Considering the yield of oil in relation to the protein content in the seeds one finds a fairly definite inverse relationship. It will be seen from table II that in some cases the protein content of the seeds decreased upon increase in oil content. This correlation bears out the findings of Schmalfuss (9) regarding the relation between nitrogen fraction and oil content.

Summary

In this study a pure line of Bison flax was grown to maturity in selected Shive's type I, three-salt nutrient solutions using a constant renewal technique. Temperature, humidity, and light conditions under which the plants grew were recorded. The data obtained on plants grown under the conditions of this experiment indicate that:

1. Final growth in height of top is relatively constant over a wide range of nutrient salt proportions.

2. The presence of fairly large quantities of KH₂PO₄ in the nutrient solutions resulted in relatively early maturity, the date of maturity being modified by the nitrogen content of the nutrient solution. The time of maturity seemed to be in inverse proportion to the amount of nitrogen present.

3. Relatively greater amounts of oven-dry material were obtained with nutrient solutions high in KH₂PO₄. The other constituent salts of the
various nutrient solutions showed no uniform relation to the oven-dry weight of plant material produced.

4. Generally the greatest seed production occurred in solutions containing the higher proportions of KH$_2$PO$_4$; other nutrient constituents show no definite relation to this criterion. The heaviest seed was obtained from plants which grew on nutrients relatively high in the potassium acid phosphate salt.

5. There is no marked variation in quantity of oil produced when the composition of the nutrient solution is varied over a rather wide range.

6. Oils from seeds produced in cultures relatively high in KH$_2$PO$_4$ have higher iodine numbers than those from cultures low in this salt.

7. The production of plants grown on nutrient solutions in constantly renewed solution cultures exceeded production of field-grown plants in the following respects: height of top, weight of seed, average protein content of seed, and iodine number of oil. The oil yields in the experimental plants about equal average oil yields of field grown plants.

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