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Simplex Optimization of Reaction Yields

Abstract. *The sequential simplex algorithm, an efficient optimization strategy, rapidly improved reaction yield as a function of time and temperature in the synthesis of* π-$C_5H_5Mo(CO)_2CSN(CH_3)_2$. *The work demonstrates the feasibility and efficiency of the simplex design and suggests its application and usefulness in other syntheses.*

Recent awareness of the finite character of both material and energy resources has stimulated a renewed interest in the optimization of reaction yields. We report here the successful application of an efficient optimization strategy that could have broad applicability in the development of synthetic chemical methods.

The development of reliable, high-yield chemical syntheses requires three efforts: (i) producing some yield of the desired compound; (ii) improving the yield; and (iii) understanding the effects of the various experimental factors (variables) upon the yield. Synthetic chemists probably devote most of their time to the definition and discovery of synthetic routes that produce the desired compound in some yield. The systematic improvement and understanding of yields are less frequently investigated, although both are often important. For example, the overall yield of a multistep synthesis is a multiplicative function of the yields at each step; improving intermediate yields is usually necessary to produce an acceptable yield of final product. Understanding the effects of the various experimental factors upon the reaction yield allows the investigator to evaluate and specify tolerances (*1*) within which each factor must be held so that the stated yield can be reliably reproduced in other laboratories.

A common optimization strategy is the one-factor-at-a-time method (*2*) in which each factor is optimized in turn while all others are held constant. Although this method is still popular (*3*), it has been shown (*4, 5*) that by this means it is impossible to find an existing optimum in many chemical systems because of ridges in the response surface. Evolutionary operation (EVOP) strategies (*6*) will succeed in the presence of ridges, but the number of experiments required is large: if k full factorial designs for n factors at m levels are needed to reach the optimum, a minimum of $k(m^n)$ experiments is required.

The sequential simplex (*7*) method of Spendley *et al.* (*8*) is a pattern search technique that requires only $n + 1$ experiments for the initial design and only one additional experiment per move. Thus, if k is the number of simplexes needed to reach the optimum, a minimum of $k + n + 1$ experiments is required. A modification by Nelder and Mead (*9*) of the original simplex algorithm allows expansions along directions that are favorable and contractions along directions that are unfavorable (*10*). The logical and mathematical details of the simplex algorithms have been discussed elsewhere (*5, 9, 11*).

The feasibility of reaction yield optimization by the simplex method is demonstrated here for the synthesis of the known compound π-$C_5H_5Mo(CO)_2CSN(CH_3)_2$ (*12*). The reaction was carried out in two steps as follows:

$$C_5H_5Na + Mo(CO)_6 \xrightarrow[t_1,\, 65°C]{THF} \pi\text{-}C_5H_5Mo(CO)_3Na + 3CO \quad (1)$$

$$\pi\text{-}C_5H_5Mo(CO)_3Na + ClCSN(CH_3)_2 \xrightarrow[22\ hr,\ T_2]{THF} \pi\text{-}C_5H_5Mo(CO)_2CSN(CH_3)_2 + CO + NaCl \quad (2)$$

where THF is tetrahydrofuran. We have investigated the effects of two potentially significant factors on reaction yield: the reaction time t_1 of Eq. 1 and the reaction temperature T_2 of Eq. 2. All other factors were held constant (*13*). The yield at each set of reaction conditions was determined by infrared spectroscopy.

The progress of the simplex toward improved reaction yield is illustrated in Fig. 1. The initial simplex (yields of 16, 74, and 82 percent) is located in a region previously investigated (*12*). Univariate mapping studies were carried out to investigate the individual effects of t_1 and T_2 in the region of the best vertex (yield, 93 percent). These results are shown in the inset in Fig. 1.

It is apparent from Fig. 1 that there is a general trend toward improved yield with increasing t_1. The slight decrease in yield above $t_1 \approx 5$ hours in the mapping study is not statistically significant. A much more dramatic effect is caused by T_2. Up to ~ 55°C, yield is improved by increasing T_2; however, above this temperature as the boiling point (b.p.) of the solvent is approached, yield decreases sharply.

This preliminary study has shown the feasibility of using a simple optimization algorithm to improve the yields of chemical syntheses. The understanding of the behavior of the reactions provided by these

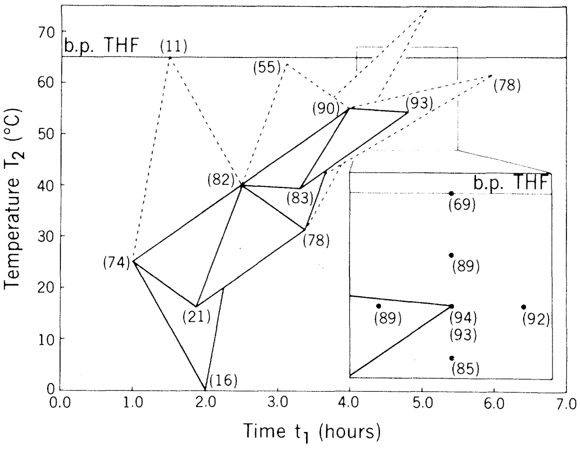

Fig. 1. Simplex improvement of yield as a function of time and temperature. Solid lines indicate simplex movement. Dashed lines show the locations of vertices generated in failed reflections and expansions. Parenthetical numbers beside the vertices are percentage yields.

experiments and the univariate mapping allows not only the determination of the best reaction conditions but also the specification of tolerances (*1*) within which the factors must be held to achieve this high yield. In particular, the sensitivity of reaction yield to T_2 suggests to us the possibility that other reactions might also exhibit a marked temperature optimum which does not necessarily coincide with the boiling point of the solvent (a choice commonly employed for reasons of convenience). A more detailed study of this and other reactions (including catalytic systems), investigating the effects of more than two factors, is in progress.

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10. Given $n + 1$ simplex vertices represented as n-dimensional vectors $\boldsymbol{W}, \boldsymbol{N}, \ldots, \boldsymbol{B}$ ranked in order of response S_W (worst), $S_N, \ldots, S_B$ (best) and centroid $\bar{\boldsymbol{P}} = \frac{1}{n}(\boldsymbol{N} + \ldots + \boldsymbol{B})$, eliminate $\boldsymbol{W}$ and evaluate the response S_R at $\boldsymbol{R} = \bar{\boldsymbol{P}} + (\bar{\boldsymbol{P}} - \boldsymbol{W})$. If $S_N \leq S_R \leq S_B$, retain $\boldsymbol{R}$. If $S_R > S_B$, evaluate S_E at $\boldsymbol{E} = \bar{\boldsymbol{P}} + 2(\bar{\boldsymbol{P}} - \boldsymbol{W})$: if $S_E > S_B$, retain $\boldsymbol{E}$; if $S_E \leq S_B$, retain $\boldsymbol{R}$. If $S_R < S_N$: if $S_R \geq S_W$, evaluate S_{C_r} at and retain $\boldsymbol{C}_r = \bar{\boldsymbol{P}} + \frac{1}{2}(\bar{\boldsymbol{P}} - \boldsymbol{W})$; if $S_R < S_W$, evaluate S_{C_w} at and retain $\boldsymbol{C}_w = \bar{\boldsymbol{P}} - \frac{1}{2}(\bar{\boldsymbol{P}} - \boldsymbol{W})$. If S_{C_r} or $S_{C_w} < S_N$, set $S_W = S_N$, $\boldsymbol{W} = \boldsymbol{N}$, $S_N = S_{C_r}$ or S_{C_w}, and $\boldsymbol{N} = \boldsymbol{C}_r$ or $\boldsymbol{C}_w$. See C. W. Lowe [*Trans. Inst. Chem. Eng.* **42**, T334 (1964)] for an example of a calculation work sheet.
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13. Reactions were carried out as follows: 15 ml of THF and 4.40 ml of a 0.50*M* solution of C_5H_5Na in THF were added to 2.00 mmole (0.528 g) of $Mo(CO)_6$. The mixture was refluxed (65°C) for t_1 and then cooled to T_2; 4.40 ml of 0.50*M* $ClCSN(CH_3)_2$ in THF were then added, and the mixture was stirred for 22 hours in a water bath held at T_2. The reaction yield was determined by adding 100 ml of THF and a weighed sample of benzonitrile (about 3.0 g) to the reaction mixture; the infrared spectrum of the solution was determined on a spectrophotometer (Perkin-Elmer model 467), and the height of the absorption maximum of the product (1841 cm^{-1}) was compared to that of benzonitrile (2245 cm^{-1}).
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Drosophila Hybrids in Nature: Proof of Gene Exchange Between Sympatric Species

Abstract. *Genetic studies of two closely related endemic Hawaiian species show that in one area of sympatry about 2 percent of the naturally occurring individuals are hybrids. More than 20 times this many would be expected if the population consisted of a single panmictic unit. Despite hybridization, natural selection appears to maintain the essential integrity of each separate gene pool.*

In the Kahuku area, near the south end of the island of Hawaii, natural interspecific hybridization has been detected between a pair of sympatric endemic species. This circumstance is very unusual for the genus, the other species of which tend to have isolated gene pools which coexist in nature without evidence of gene exchange (*1*). Nevertheless, laboratory crosses between sympatric species are frequently possible and sometimes one or both sexes of F_1 hybrids are fertile (*2*). Accordingly, if reproductive isolation became weak, the potential for gene exchange would exist.

In an extensive literature on *Drosophila* species extending over 50 years, only two cases suggest the possibility of introgression in nature. In both, laboratory crosses give fertile F_1 females and sterile F_1 males. In *D. pseudoobscura* and *D. persimilis*, close genetic scrutiny of the progeny of 27,099 wild-caught flies from sympatric areas yielded four cases of F_1 hybridization (*3*). In each case, a wild female of one species had mated with a male of the other species. Considerable doubt exists, however, as to whether the interspecific matings observed actually took place in nature; since the wild flies were not separated at capture, the interspecific mating could have occurred in the collecting vials on the way to the laboratory. Only a single instance of a backcross hybrid was detected. In *D. metzii* and *D. pellewae*, introgression in nature may be strongly inferred from a face color polymorphism, but critical genetic proof of hybridization in nature is lacking (*4*). We report here a case of hybridization in which one F_1 and three backcross hybrid individuals have been unequivocally recognized in nature.

Drosophila setosimentum and *D. ochrobasis* are near-sibling species endemic to the island of Hawaii (*5*). *Drosophila setosimentum* occurs in rain forests between 600 and 1600 m altitude. At five widely separated locations, all above 1000 m, it is accompanied by *D. ochrobasis*. Both species are rare and local, being largely confined to kipukas (islands of vegetation isolated in the midst of newer lava flows) or collapsed lava tubes where their rare host plants grow. Although no reliable means exists for the separation of females on morphological grounds, males of the two species are easily distinguished by a secondary sexual difference in wing pattern.

Wild-caught females were separated from males at capture. In the laboratory, each female was placed in a separate culture vial and allowed to produce offspring. Such a culture (an isofemale line) usually yields F_1 males conforming exclusively to either one species or the other. From each isoline, seven (sometimes fewer) F_1 larvae were taken, and an acetoorcein squash preparation of the salivary gland chromosomes from each was prepared. From each smear the banding order of all five major chromosome arms (X, 2, 3, 4, and 5) was read directly along the full length of the chromosome, with the use of spaced landmark areas to confirm the sequences. The banding orders of all paired homologs were thereby determined. Inverted sections were read in either homozygous or heterozygous state. The sequence of the hemizygous X in each male larva was also determined.

These cytological data permit inferences about the state of the natural population from which the wild flies were drawn. Except in the Kahuku Ranch population, to be described later, all the F_1 test larvae (195 female and 145 male larvae) examined from 56 wild *D. ochrobasis* females have shown a uniform sequence in chromosome X (*ochrobasis* standard X). In addition, all larvae are homozygous for a chromosome 2 inversion (2k), and all lack an inversion (2l) near the opposite end of the same chromosome. This latter inversion is fixed in all but a North Kona, Hawaii, population of *D. setosimentum*. In addition, the commonest gene order of chromosome 4 of *D. ochrobasis* differs from the standard 4th chromosome of *D. setosimentum* by six inversions. These are spread in a roughly tandem manner over the length of the chromosome.

Similarly, 154 female and 164 male test larvae have been recognized from sympatric areas as the progeny of 54 wild *D. setosimentum* flies. In allopatric areas, 221