1-Benzyl-5-bromo-3-methyl-2-pyridone (9). A solution of 1.0 g (5.02 mmol) of **8** in 10 ml of dry benzene was placed in a dry, nitrogen-filled flask. To the solution was added 0.90 g (5.02 mmol) of NBS and the mixture was heated at 90 °C for 50 min. The benzene was removed under reduced pressure, 25 ml of carbon tetrachloride was added to the residue, and the resulting mixture was filtered. The filter cake was washed with 25 ml of carbon tetrachloride and the filtrate was concentrated under reduced pressure leaving an orange oil as residue, the NMR of which showed less than 10% of **10**. The oil crystallized on cooling, and trituration with 10 ml of anhydrous ether gave 0.95 g (68%) of crude **9** as a white solid, mp 86.5–89 °C. The solid was recrystallized twice from ether to give an analytical sample of **9**: mp 96.5–97.5 °C; NMR ($CDCl_3$) δ 7.04–7.60 (m, including s at 7.23, 7 H total), 5.02 (s, 2 H), 2.12 (s, 3 H).

Anal. Calcd for $C_{13}H_{12}BrNO$: C, 56.14; H, 4.35; N, 5.04. Found: C, 55.98; H, 4.56; N, 5.00.

The reaction in 10 ml of carbon tetrachloride gave identical results. The product crystallized and NMR of the crude solid showed about 10% of **10**.

1-Benzyl-3-bromomethyl-2-pyridone (10). In a 100-ml flask equipped with a reflux condenser and a drying tube was placed a solution of 1.0 g (5.02 mmol) of **8** in 75 ml of dry carbon tetrachloride. To the solution was added 0.90 g (5.02 mmol) of NBS and 0.1 g of dibenzoyl peroxide. The mixture was heated under reflux with a 100-W lamp for 4 h.[12] After this time the mixture was cooled and filtered, and the solvent was removed to afford a yellow oil which solidified on cooling under 15 ml of anhydrous ether to give 1.0 g (72%) of crude **10**, mp 86–90 °C, the NMR spectrum of which showed no product of ring bromination, **9**. The solid was recrystallized twice from ether to give an analytical sample of **10**: mp 101–101.5 °C; NMR ($CDCl_3$) δ 7.30–7.60 (m, including s at 7.34, 7 H total), 6.17 (t, 1 H), 5.17 (s, 2 H), 4.47 (s, 2 H).

Anal. Calcd for $C_{13}H_{12}BrNO$: C, 56.14; H, 4.35; N, 5.04. Found: C, 56.23; H, 4.43; N, 4.94.

The reaction was repeated using only 10 ml of carbon tetrachloride and the NMR of the crude solid showed the presence of 20–30% of **9**.

3-Bromomethyl-1-methyl-2-pyridone (6). In a dry nitrogen-filled 250-ml flask was placed a solution of 0.83 g (6.7 mmol) of **5** in 100 ml of dry carbon tetrachloride. To the solution was added 1.19 g (6.7 mmol) of purified NBS[9] and 0.15 g of dibenzoyl peroxide. The mixture was heated under reflux for 1 hr. After this time the mixture was cooled and filtered, and the solvent removed. The solid residue[10] was stirred under 10 ml of anhydrous ether and was removed by filtration to afford 0.85 g (63%) of **6** as tan crystals, mp 86–89 °C. The product was recrystallized twice from benzene to give an analytical sample of **6**: mp 101–101.5 °C (lit.[7] mp 98–99 °C); NMR ($CDCl_3$) δ 7.67 (m, 2 H), 6.31 (t, 1 H), 4.58 (s, 2 H), 3.65 (s, 3 H).

Anal. Calcd for C_7H_8BrNO: C, 41.61; H, 3.99; N, 6.93. Found: C, 41.68; H, 4.06; N, 6.83.

5-Bromo-1,3-dimethyl-2-pyridone (7). A solution of 0.83 g (6.7 mmol) of **5** in 12 ml of dry carbon tetrachloride was placed in a dry nitrogen-filled flask. To the solution was added 1.18 g (6.6 mmol) of purified NBS[9] and the mixture was heated under reflux for 30 min. After this time 25 ml of carbon tetrachloride was added; the mixture was cooled and filtered; and the solvent was removed to afford 1.28 g (96%) of **7** as a light-yellow solid,[11] mp 98–101 °C. Recrystallization of the product from petroleum ether (bp 30–60 °C) gave fluffy, white crystals: mp 105–106 °C (lit.[3] mp 106–107 °C); NMR ($CDCl_3$) δ 7.30–7.55 (m, 2 H), 3.61 (s, 3 H), 2.20 (s, 3 H).

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Registry No.—**5**, 6456-92-4; **6**, 58802-10-1; **7**, 51417-13-1; **8**, 58802-11-2; **9**, 58802-12-3; **10**, 58802-13-4; 3-methyl-2-pyridone, 1003-56-1; benzyl chloride, 100-44-7; methyl iodide, 74-87-3; *N*-bromosuccinimide, 128-08-5.

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(9) The NBS was purified by recrystallization from ten times its weight of water and drying under vacuum overnight (mp 182.5–184 °C).
(10) The residue contained less than 10% of compounds **5** and **7**, combined, by NMR.
(11) The product contained only a trace (<2%) of compounds **5** and **6** by NMR.
(12) The manner and the time of heating after 50 min is not critical.

Biological Probes. 3. Methods for Carbon-4 and Carbon-5 Labeling in Nicotinamide

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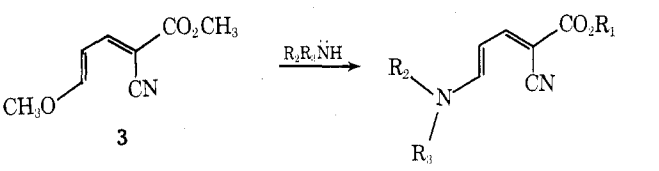
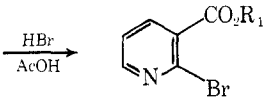
Increased interest in nonradioactive labels for use as general biological probes had led us to develop efficient methods for labeling the nicotinamide (**1**) portion of NAD^+ (**2**).[1-3] We have described facile pyridine syntheses in which nicotinamide can be labeled (^{13}C, ^2H, ^{15}N) at the 1, 2, 3, 6, and carbonyl positions and then be biosynthetically incorporated into the coenzyme NAD^+.[4] However, these methods were not useful for labeling the 4 position of the nicotinamide ring, the site at which biological oxidation–reduction occurs in NAD^+. We now wish to report an efficient, high-yield procedure for label incorporation (^{13}C, ^2H) at the 4 and also the 5 position of nicotinamide (**1**).

Prior experience with diene **3** as a labeled pyridine precursor suggested an attractive synthetic route to **1**. Our initial studies focused on modification of diene **3** with designs on making this general type of synthon more accessible from lower molecular weight, labeled starting materials. Specifically, diene **3** readily undergoes addition (1,6) of amines with loss of methanol forming butadienamines, such as **4**. These conjugated enamines (**4**), analogous to diene **3**, undergo acid-catalyzed (HBr/AcOH) cyclization to 2-bromonicotinate **5** in high yield.



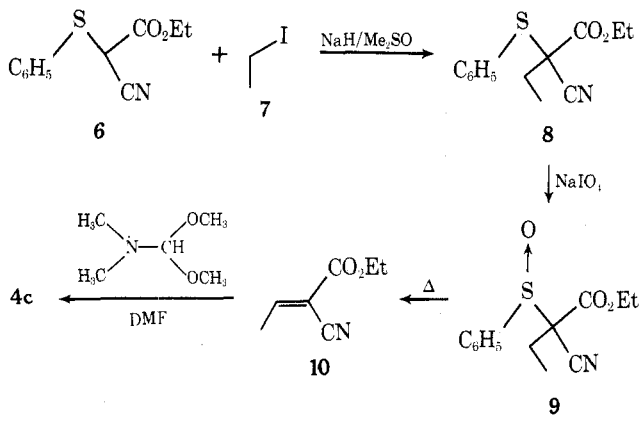
3 **4a**, $R_1 = R_2 = Me$; $R_3 = C_6H_5$
b, $R_1 = R_2 = R_3 = Me$
c, $R_1 = Et$; $R_2 = R_3 = Me$



5, R = Me or Et

Therefore, several routes to **3** or **4** were investigated with our labeling goals in mind resulting in the preparation of enamine **4c** as shown in Scheme I.

Scheme I



Sulfenyl cyanoacetate **6,** prepared by treating *N,N*-diethylbenzenesulfenamide[5] with cyanoacetate, can be alkylated using NaH in Me_2SO with specifically labeled ethyl iodide (**7**) affording the cyanobutyrate **8** in 84% yield. Using extended reaction times (48 h) and finely powdered $NaIO_4$,[7] cyanobutyrate **8** was smoothly oxidized to sulfoxide[8] **9** which was subjected in its crude form to thermolysis in refluxing toluene yielding ethylidene **10** in 89% yield. Transformation of **10** to enamine **4c** was achieved through an amide acetal condensation. Ethyl ethylidene cyanoacetate (**10**) when treated with *N,N*-dimethylformamide dimethyl acetal in warm DMF affords **4c** in 81% yield (60% from **6**). As cited above, **4c** undergoes facile HBr-catalyzed cyclization to 2-bromonicotinate **5** in 95% yield. Catalytic reduction of **5** followed by treatment with aqueous ammonia leads to nicotinamide (**1,** 45% yield from **6**) as described earlier.

Ethyl iodide was not our initial choice as a labeling unit. However, the reproducibly high yields obtainable from alkylation of **6** followed by subsequent formation of ethylidene **10** (75% from **6**) proved superior to alternatives such as a Knoevenagel condensation with cyanoacetate and acetaldehyde. Preliminary studies indicated that such reactions are at best low yield conversions to **10,** and the potential expense of using specifically labeled acetaldehyde is also prohibitive.

Through this and earlier studies, methods now exist for the preparation of specifically labeled nicotinamide from simple labeled precursors through convenient, high-yield reactions. In addition, such reactions would appear to be general and usable for the preparation of other important pyridine systems.[10]

Experimental Section[9]

Ethyl Phenylthiocyanoacetate (6)[6]. A mixture of *N,N*-diethylbenzenesulfenamide (24.54 g, 0.134 mol) and ethyl cyanoacetate (15.17 g, 0.134 mol) was stirred in methylene chloride (150 ml) at room temperature for 5 h. Removal of the volatiles at reduced pressure gave pale yellow crystals which were then stirred in a mixture of 10% hydrochloric acid (300 ml) and benzene (300 ml) at room temperature for 1 h. The benzene layer was separated and the solvent removed at reduced pressure to give 27.57 g of a pale yellow oil. Fractional distillation (120 °C, 2.5 mmHg) afforded 20.03 g (67%) of ethyl phenylthiocyanoacetate: ^1H NMR δ_{CDCl_3} (Me_4Si) 7.76–7.06 (m, 5 H, C_6H_5), 4.34 (s, 1 H, C_2 H), 4.16 (q, J = 7 Hz, 2 H, $-OCH_2$), 1.20 (t, J = 8 Hz, 3 H, $-CH_3$); ir (film) 2300, 1740, 1580 cm^{-1}; TLC (silica gel 1:1:1 $CH_3OH/EtOAc/CH_2Cl_2$) R_f 0.73; *m/e* 221.

Ethyl 2-Phenylthio-2-cyanobutyrate (8). Phenylthiocyanoacetate **6** (5 g, 22.7 mmol) was added to NaH (57% dispersion, washed once with hexane, 0.96 g, 1 molar equiv) in Me_2SO at 0 °C. The mixture was then allowed to equilibrate at room temperature for 20 min. Ethyl iodide (3.53 g, 22.7 mmol) was added and the mixture stirred for 6 h, diluted with H_2O (100 ml), and extracted with ether/hexane (3:1, 4 × 75 ml). The organic extracts were combined, washed with brine (100 ml), and dried (Na_2SO_4). Distillation (Kugelrohr oven, 0.5 mmHg, 120–135 °C) afforded 4.76 g (84%) of butyrate **8:** ir (film) 2250, 1740, 1580 cm^{-1}; ^1H NMR δ_{CDCl_3} (Me_4Si) 7.79–7.08 (m, 5 H, C_6H_5), 4.06 (q, 2 H, J = 7 Hz, $-OCH_2-$), 2.41–1.74 (m, 2 H, $-CH_2-$), 1.34–0.9 (m, 6 H, $-CH_3$); TLC (silica gel, $CHCl_3$) R_f 0.54; *m/e* 249. Anal. Calcd for $C_{13}H_{15}NO_2S$: C, 62.64; H, 6.07. Found: C, 62.66; H, 6.02.

Ethyl 2-Cyano-2-butenoate (10). A saturated aqueous solution of finely powdered $NaIO_4$ (15 ml) was added to phenylthiocyanobutyrate **8** in MeOH (30 ml) at 0 °C. The mixture was stirred mechanically at 0 °C for 10 min and then at room temperature for 48 h. The mixture was diluted with H_2O (50 ml), extracted with $CHCl_3$ (4 × 75 ml), washed (NaCl), and dried ($MgSO_4$). The volatiles were removed at reduced pressure to afford 1.04 g of a yellow oil, which was dissolved in toluene (40 ml), stirred, and heated under reflux overnight. The mixture was concentrated at atmospheric pressure and distillation (Kugelrohr oven, 1.0 mmHg, 80 °C) afforded 0.45 g (89%) of butenoate **10:** ir (film) 2275, 1735, 1635 cm^{-1}; ^1H NMR δ_{CDCl_3} (Me_4Si) 7.78 (q, J = 8 Hz, 1 H, C_3 H), 4.38 (q, J = 7 Hz, 2 H, $-OCH_2$), 2.26 (d, J = 8 Hz, 3 H, $-C_4H_3$), 1.38 (t, J = 7 Hz, $-CH_3$); TLC (silica gel, $CHCl_3$) R_f 0.37 (0.54 starting material); *m/e* 139. Anal. Calcd for $C_7H_9NO_2$: C, 60.42; H, 6.52. Found: C, 60.39; 46.52.

Ethyl 5-(*N,N*-dimethylamino)-2-cyano-2,4-pentadienoate (4c). *N,N*-Dimethylformamide dimethyl acetal (0.31 g, 2.6 mmol) was added to ethyl 2-cyano-2-butenoate (0.35 g, 2.5 mmol) in DMF (1 ml). The mixture was stirred and heated at 75 °C for 5 h, cooled to room temperature, poured into benzene (20 ml), and washed with 1 N HCl (3 × 25 ml). The benzene solution was dried (Na_2SO_4) and the soluent removed at reduced pressure. Distillation (Kugelrohr oven, 140–150 °C, 2.5 mmHg) afforded 0.31 g (81%, mp 125–127 °C) of enamine **4c:** ir ($CHCl_3$) 2220, 1700, 1620, 1560 cm^{-1}; ^1H NMR δ_{CDCl_3} (Me_4Si) 7.78 (d, J = 13 Hz, 1 H, C_5 H), 7.10 (d, J = 13 Hz, 1 H, C_3H), 5.59 (t, J = 13 Hz, 1 H, C_4 H), 4.26 (q, J = 7 Hz, 2 H, $-OCH_2-$), 3.11 (s, 6 H, $-NCH_3$), 1.32 (t, J = 7 Hz, 3 H, $-CH_3$); λ_{max} (EtOH) 381 nm; TLC (silica gel, $CHCl_3$) R_f 0.08; *m/e* 194. Anal. Calcd for $C_{10}H_{14}N_2O_2$: C, 61.83; H, 7.27. Found: C, 61.88; H, 7.25.

Ethyl 2-Bromonicotinate (5). Enamine **4c** (1 g, 3.49 mmol) was dissolved in 5 ml of acetic acid. An acetic acid solution of HBr (10 ml, saturated at 0 °C) was added dropwise to enamine **4c** while maintaining the reaction at 45 °C. After addition of the HBr solution, the temperature was raised to 55 °C and the mixture was allowed to stir for 2 h. The dark solution was cooled, poured into water, and neutralized by Na_2CO_3. The aqueous solution was extracted with CH_2Cl_2 (3 × 150 ml). The CH_2Cl_2 extracts were combined, washed once with H_2O (100 ml), and dried (Na_2SO_4). Evaporation of the volatiles at reduced pressure gave a dark oil. Distillation (Kugelrohr oven, 0.5 mmHg, 110–125 °C) afforded 0.76 g (95%) of ethyl 2-bromonicotinate: ^1H NMR δ_{CDCl_3} (Me_4Si) 8.47 (dd, $J_{6,4}$ = 2, $J_{6,5}$ = 5 Hz, 1 H, C_6H), 8.07 (dd, $J_{4,5}$ = 8.5, $J_{4,6}$ = 2 Hz, 1 H, C_4 H), 7.40 (dd, $J_{4,5}$ = 8.5, $J_{5,6}$ = 5 Hz, 1 H, C_5 H), 4.42 (q, J = 7 Hz, 2 H, $-OCH_2-$), 1.43 (t, J = 7 Hz, 3 H, $-CH_3$); ir ($CHCl_3$) 1735, 1580 cm^{-1}; TLC (silica gel, $CHCl_3$) R_f 0.24 (starting material, 0.08); *m/e* 230. Anal. Calcd for $C_8H_8NO_2Br$: C, 41.74: H, 3.51. Found: C, 41.70; H, 3.54.

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Registry No.—**4c,** 51513-16-7; **5** (R = Et), 53087-78-8; **6,** 58734-93-3; **8,** 58734-94-4; **10,** 686-33-9; *N,N*-diethylbenzenesulfenamide, 6667-19-2; ethyl cyanoacetate, 105-56-6; *N,N*-dimethylformamide dimethyl acetal, 4637-24-5.

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(8) Presence of sulfoxide [Ph–S(→O)–R] was detected by infrared absorption at 705 cm^{-1}.
(9) Structural assignments of all compounds are based on ir, ^1H NMR, uv, mass spectra, analysis, and conversion to previously reported organic compounds.
(10) A variety of 2-substituted nicotinamide compounds exhibit anti-inflammatory activity.