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Precursors of Chicken Flavor. II. Identification of Key Flavor Precursors Using Sensory Methods

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Sensory evaluation was used to identify flavor precursors that are critical for flavor development in cooked chicken. Among the potential flavor precursors studied (thiamin, inosine 5′-monophosphate, ribose, ribose-5-phosphate, glucose, and glucose-6-phosphate), ribose appears most important for chicken aroma. An elevated concentration (added or natural) of only 2−4-fold the natural concentration gives an increase in the selected aroma and flavor attributes of cooked chicken meat. Assessment of the volatile odor compounds by gas chromatography−odor assessment and gas chromatography−mass spectrometry showed that ribose increased odors described as "roasted" and "chicken" and that the changes in odor due to additional ribose are probably caused by elevated concentrations of compounds such as 2-furanmethanethiol, 2-methyl-3-furanthiol, and 3-methylthiopropanal.

KEYWORDS: Chicken; ribose; ribose-5-phosphate; IMP; thiamin; flavor precursors; 2-furanmethanethiol; 2-methyl-3-furanthiol; 3-methylthiopropanal

INTRODUCTION

Meat flavor develops during cooking by complex reactions between natural components present in raw meat. These precursors may include reducing and phosphorylated sugars, amino acids, thiamin, and lipids (*1*, *2*). The cooking of meat generates many hundreds of volatile compounds, but relatively few make a key contribution to the odor and flavor of cooked meat. Many of these key odor compounds in chicken have been identified (*2*), and often, mechanisms for their formation have been suggested.

Many flavor compounds may be formed by two or more possible mechanisms. For example, furanthiols, which make an important contribution to meaty odors in most meats, may be formed by the reaction of cysteine and ribose (*3*, *4*), cysteine, and inosine 5′-monophosphate (IMP) (*5*) or the degradation of thiamin (*6*). Therefore, the relative importance of each precursor for flavor generation in cooked chicken meat is still unclear.

The question of which of the many possible precursors is critical for flavor formation has been addressed in lamb, beef, and pork by monitoring the changes in volatile compounds and/or sensory quality following the addition of precursors into meat (*7*−*10*). These studies have suggested that relatively small added quantities of sugars and nucleotides (e.g., 2- or 4-fold the natural concentration) may be sufficient to increase meaty and roasted odor. Preliminary studies (Farmer and Nolan, unpublished data) suggested that similar effects may occur in chicken.

This study aims to determine the importance of individual precursors for chicken odor and flavor. The work has concentrated on those precursors believed to affect the Maillard and related flavor-forming reactions in meat. Previous papers in this series report the natural concentrations for ribonucleotides, thiamin and amino acids (*11*), and sugars and their phosphates (*12*). While this information was not available for the first experiment reported herein, subsequent experiments made use of this analytical data to allow the addition of approximately 2−5-fold the natural concentrations into raw chicken, prior to analysis of the odor or flavor by sensory or instrumental methods. In addition, the results of a preliminary study on the flavor differences between chicken muscles with differing natural concentrations of precursors are reported.

EXPERIMENTAL PROCEDURES

Materials. D-Ribose-5-phosphate disodium salt (R5P), D-glucose-6-phosphate disodium salt hydrate (G6P), D-ribose, D-glucose, thiamin hydrochloride, L-cysteine, and IMP were purchased from Sigma (Poole, United Kingdom). "Food grade" D-ribose was obtained from Aldrich Flavors and Fragrances (Poole, United Kingdom). Glucose (Dextrose Powder) from Thornton & Ross Ltd. (Huddersfield, United Kingdom) was used as a source of food grade glucose. A mixture (51:49%) of food grade IMP and guanosine 5′-monophosphate (GMP) was provided as a gift from Quest International (Menstrie, Scotland). Authentic flavor compounds were purchased from Sigma-Aldrich except for 2-methyl-3-methyldithiofuran, which was prepared in a mixture of 2-methyl-3-furanthiol and dimethyldisulfide (Sigma-Aldrich). Distilled water was double distilled in an Aquatron A4D still (J. Bibby Sterilin Ltd., Stone, United Kingdom) fed by tap water prefiltered through a charcoal filter (Fistreem, Loughborough, United Kingdom).

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Table 1. List of Compounds and Concentrations Used in Experiments 1–3

compound	natural concn (lit)[a] (mg 100 g^{-1})	ref[b]	natural concn (own analyses)[c] (mg 100 g^{-1})	ref	expt 1 (mg added to 100 g of meat)[d]	expt 2 (mg added to 100 g of meat)	expt 3 (mg added to 100 g of meat)
IMP	330	*29*	75	*11*	400	75, 150	190
ribose	1–14	*30*, *31*	25	*12*	150	50, 100	100
R5P			14	*12*		56, 100	
cysteine	28	*31*	0.03	*11*	125	50	
cysteine + ribose	28 + 14		0.03 + 25	*12*	125 + 150	50 + 100	
glucose	188	*31*	40	*12*	180	80	100
G6P			17	*12*	300	100	
thiamin	0.15	*32*, *33*	0.2	*11*	340	100, 0.5	

[a] Natural concentration of these compounds in chicken meat, as reported in the literature. [b] References. [c] Natural concentration of these compounds in chicken meat, as determined in chicken from the same source and at the same time after slaughter as that used for these experiments (*11*, *12*). [d] Amount added (mg) to 100 g of ground meat in 10 mL of distilled water (expt 1 and 2) or tap water (expt 3).

Chicken. Fresh chicken breast fillets (*M. pectoralis major*) without skin or bone from "standard" chickens were supplied by Moy Park Ltd. (Northern Ireland) for all sensory studies. The chicken meat was transported at 4 °C, and each batch (5 kg) was ground (minced) within 2 h thereafter. The ground chicken breast was thoroughly mixed, divided into small portions (1 kg), and vacuum packed in polyethylene vacuum bags (Somerville Packing, Lisburn, Northern Ireland). The packs were frozen on the same day, approximately 4−5 h after slaughter, and held at −20 °C until use.

The day before a sensory panel, the required quantity of frozen minced chicken was thawed to room temperature (20 °C), and requisite quantities of precursor(s) for each of the experiments described below were added in 10 mL of water for each 100 g of ground chicken. The treated chicken meat was then homogenized using a food processor (Robot chef 2, Robot-Coupe, Vincennes, France). A control sample was prepared by adding water instead of precursor solution (10 mL 100 g^{-1} minced meat). In samples for sensory analysis of odors only, distilled water was used for the preparation of solutions of added precursors, while for samples destined for flavor analysis, tap water was used together with "food use" precursors.

Treated and control meat samples were kept in 3 L beakers at 4 °C overnight to allow equilibration. The following day, meat samples for sensory analysis were divided into 20 g samples and transferred into 100 mL beakers, covered with aluminum foil, and cooked at 120 °C for 30 min in a fan-assisted oven (Falcon E1102, Glynwed Appliances, Laberth, United Kingdom). Samples for volatile collection and instrumental analysis were cooked under the same conditions in a Plus II oven (Sanyo Gallenkamp plc., Loughborough, United Kingdom).

Sensory Experiments. A series of sensory experiments were conducted to screen a number of potential precursors to determine the most important sensory attributes affected and to identify those precursors most important for these changes.

Experiment 1: Effect of Selected Precursors on Different Sensory Attributes of Cooked Chicken Meat. The panelists developed and agreed profiling terms and descriptors for the odors of ground chicken breast samples to which a range of selected compounds had been added prior to cooking (**Table 1**). The effect of added IMP, ribose, glucose, G6P, cysteine, ribose + cysteine, and thiamin on the odor quality of cooked chicken was studied using sensory profiling. The quantities of these substances added to chicken were estimated from literature reports, as shown in **Table 1**. Eight replicate samples were assessed for each treatment by six trained panelists on a 10 cm line scale, anchored at 5 and 95 mm by terms such as "not sweet" and "very sweet". A computerized sensory data collection system (PSA 3, version 2.07, Oliemans Punter and Partners bv, Utrecht, The Netherlands) was used for this and subsequent sensory experiments.

Experiment 2: Effect of Low Added Concentrations of Precursors on Odor. A paired comparison test was employed to compare chicken samples treated with a range of compounds with control samples (**Table 1**). The concentrations used were related to those determined in chicken from the same commercial source. Thirty experienced but untrained panelists were asked to select the sample that had more "roasted aroma", "meaty aroma", "chicken aroma", or "off-odor", with intervals of 5 min between pairs of samples for each question. These attributes were chosen based on their importance, as shown in experiment 1 and previous preliminary experiments (Farmer and Nolan, unpublished data). The term off-odor was included to allow panelists to indicate if any of the precursors caused an odor not naturally expected in meat. However, interpretation of the answers to this question required care.

Experiment 3: Effect of Low Added Concentrations of Precursors on Flavor. For three of the precursors (ribose, glucose, and a mixture of IMP and GMP), the effect on the flavor of cooked chicken was investigated using food-grade precursors (**Table 1**). Thirty panelists tasted pairs of cooked ground chicken breast and selected the sample that had more "chicken flavor", "meaty flavor", "roasted flavor" and "off-flavor".

Experiment 4: Effect of Natural Variation of Ribose and Ribose Phosphate on Flavor Cooked Chicken Breast. The left breast and leg of each of 12 chickens from two different commercial sources (six of each) were analyzed for sugars and sugar phosphates by the method described previously (*12*). The remainder of these chicken carcasses were vacuum-packed and kept at −20 °C for the sensory panel, which took place 2 weeks later.

Each chicken (A−L) was categorized according to its concentration of ribose in the breast meat and, where differences in ribose concentration were small, ribose phosphate. A paired comparison test was designed to compare, first, breast and leg meat from three chickens with "low" breast concentrations of ribose (7.1−12.2 mg 100 g^{-1}) with that from three chickens containing a "high" amount of ribose (22.5−31.9 mg 100 g^{-1}), and second, chickens with "medium" (12.6−19.4 mg 100 g^{-1}) vs "high" concentration of ribose (18.8−30.4 mg 100 g^{-1}). The right breast and leg meat (combined muscles removed from thigh and drumstick) from each chicken were wrapped individually in aluminum foil and cooked at 120 °C for 30 min. For the comparison of one chicken with high ribose concentration and one with low levels, 10 panelists compared the breast meat and five of them also compared the thigh. Different sets of panelists compared the breast and thigh from the second and third pairs of chickens. These same panelists also compared chickens with medium and high ribose concentrations. Only one question was asked, "Which sample has the most roasted chicken flavor?" The low quantities of chicken meat available for each comparison meant that the numbers of panelists making these comparisons were very limited. To obtain an estimate of statistical significance, the 30 panelists comparing low with high (or medium with high) were considered together.

Experiment 5: Effect of Low Added Concentrations of Precursors on Volatile Odors by Gas Chromatography−Odor Assessment (GC-O). Chicken breast samples (control) were compared with chicken samples to which four or six times the natural quantity of ribose or R5P (100 mg 100 g^{-1}) had been added of these sugars. Ground chicken breast meat (20 g), with or without added precursor, was placed in a 100 mL Duran bottle (Davidson & Hardy Ltd., Belfast, United Kingdom), which was covered with aluminum foil and cooked at the same temperature and time as described for the sensory studies in a Plus II oven (Sanyo Gallenkamp Plc). The aluminum foil cap was then

Table 2. Effect of Added Precursors on Mean Profiling Scores for 17 Odor Descriptors for Cooked Homogenized Chicken (Experiment 1)[a]

	added compound to meat (mg 100 g^{-1})									
variable	control	IMP (400 mg)	ribose (150 mg)	thiamin (340 mg)	glucose (180 mg)	G6P (300 mg)	cysteine (120 mg)	cysteine + ribose (120 + 150 mg)	SEM	sig
strength	53.4[b]	54.4	58.6	61.0	52.9	54.2	57.0	57.3	2.10	NS[c]
roasted	30.6 ab	31.2 ab	36.7 bc	38.0 c	33.6 abc	30.4 ab	28.1 a	35.2 bc	2.10	**
chicken	29.9 ab	31.7 ab	31.1 ab	39.4 c	29.3 ab	28.2 a	35.0 bc	39.6 c	1.98	***
bloody	29.8 bc	26.9 abc	22.0 a	29.7 bc	27.32 bc	27.6 abc	32.9 c	26.5 ab	1.90	**
veg/soup	28.7 bc	24.2 ab	25.2 ab	32.4 c	23.5 ab	22.5 a	28.4 abc	29.2 bc	1.86	**
savory	30.8 ab	28.8 ab	32.0 ab	34.1 b	29.2 ab	27.8 a	27.6 a	34.7 b	1.91	*
oil/fatty	29.4	28.6	23.9	25.8	27.7	29.2	26.2	25.7	1.70	NS
rancid	22.9 bc	19.2 ab	16.3 a	20.1 ab	20.1 ab	21.0 ab	27.6 c	20.4 ab	1.96	**
buttery	22.6	24.2	23.6	22.8	21.0	21.7	20.7	22.7	1.45	NS
salty	25.6 a	30.1 a	28.9 a	31.7 a	26.5 a	26.7 a	27.6 a	45.5 b	3.68	**
sweet	21.3	25.3	21.8	24.6	23.4	24.7	21.3	21.9	1.73	NS
sour	17.2 a	15.6 a	14.8 a	19.0 a	18.3 a	16.3 a	24.9 b	19.6 a	1.58	***
fishy	15.0	14.2	13.6	15.7	17.4	14.9	16.1	17.0	1.23	NS
earthy	25.6	23.7	28.8	22.6	24.2	24.5	21.4	22.6	1.80	NS
nutty	23.0 a	24.5 a	31.2 b	24.0 a	24.9 a	23.0 a	20.1 a	21.9 a	1.70	***
woody	19.9 a	22.5 a	30.0 b	21.4 a	24.6 ab	23.2 a	20.3 a	20.7 a	1.81	**
chemical	16.6	19.5	17.9	16.1	16.4	18.3	18.5	20.7	1.51	NS

[a] Values sharing the same superscript are not significantly different by Duncan's test. [b] Values represent means sensory scores from eight replicate assessments by five panelists. [c] NS, not significant.

quickly removed, the cooked chicken meat was broken up for 10 s using a glass rod, and the Duran bottle was fitted with a screw-threaded lid with a Teflon-coated septum. The static headspace collection of volatiles from cooked chicken and the GC-O analyses were performed as described previously (*13*). The odors of eluted volatiles were described and scored on a scale of 1 (very weak) to 5 (very strong), by four experienced assessors for a period of 60 min. Each person assessed four samples in duplicate, together with duplicate blank samples, prepared using the same procedure without chicken meat. Samples were presented in a randomized order.

Experiment 6: Identification of Volatile Odors Using Gas Chromatography−Mass Spectrometry (GC-MS). Dynamic headspace concentration was used to collect a sufficient quantity of the main volatile compounds from cooked chicken meat for GC-MS analysis. Homogenized chicken breast (50 g) was placed in an Erlenmeyer flask (250 mL) covered with aluminum foil. The chicken sample was cooked as previously described. Dynamic headspace collection of volatiles from cooked chicken and the GC-MS analyses were performed as described in a previous paper (*22*). A solution of alkanes (C_8−C_{22}, approximately 20 ng μL^{-1} in ethanol) was employed as an external reference to provide linear retention indices (LRI), and an internal standard (bromobenzene, 20 ng in 1 μL) was included in each collection, as described previously (*13*).

Analyses were conducted on a Hewlett-Packard 5973 mass selective detector connected to a Hewlett-Packard 6890 GC, operated in the electron impact mode at 70 eV and an ion source temperature of 220 °C, over the range 35−450 amu. The GC program and column were as described for GC-O. Single ion monitoring (SIM) was also used to target the ions of selected compounds; 2-methyl-3-furanthiol and 2-furanmethanethiol were monitored on ions 114, 85, and 81 and 81, 114, and 53, respectively.

GC-MS analyses focused on the identification of key odor compounds, as detected by GC-O. Compounds were identified by comparison of their mass spectra, LRI, and odor description with those of authentic standards, analyzed as described above and previously (*13*), or where this was not possible, tentative identities were proposed based on comparison with details published in the literature and the NIST/Mass Spectral Database.

RESULTS

Experiment 1: Effect of Selected Precursors on Different Sensory Attributes of Cooked Chicken Meat. Table 2 shows the mean scores for 17 descriptors for the odor of cooked chicken with and without selected precursors. The addition of potential precursors caused significant increases in the intensity of roasted aroma (thiamin), chicken aroma (thiamin and cysteine + ribose), and also nutty aroma and woody aroma (ribose). Ribose also decreased rancid aroma and bloody aroma.

Table 3. Results of Paired Comparison Test of the Odor of Cooked Homogenized Chicken Breast Samples with and without Added Precursors (Experiment 2, Control vs Treated)

			odor			
precursor	mg added to 100 g of meat[a]	added concn	chicken	meaty	roasty	off
IMP	75	1×	16:14[b]	15:15	18:12	10:20
IMP	150	2×	14:16	15:15	12:18	**09:21**[c]
ribose	50	2×	**08:22**	14:16	11:19	15:15
ribose	100	4×	11:19	12:18	**08:22**	20:10
ribose	100	4×	10:20	**08:22**	10:20	19:11
cysteine	50	70×	16:14	15:15	11:19	16:14
ribose + cysteine	100 + 50	4× + 70×	11:19	12:18	**08:22**	20:10
R5P	56	4×	11:19	11:19	12:18	13:17
R5P	100	7×	14:16	13:17	15:15	18:12
glucose	80	2×	16:14	10:20	16:14	14:16
G6P	100	5×	15:15	16:14	17:13	19:11
thiamin	0.50	2×	17:13	12:18	12:18	15:15
thiamin	0.90	(4.5)×	15:15	11:19	19:11	15:15
thiamin[d]	100	450×	11:19	**09:21**	10:20	16:11

[a] Milligrams of compound added to 100 g of minced meat in 10 mL of distilled water. [b] Numbers of panelists selecting control: treated sample as possessing more of this attribute; for 30 panelists. Critical values are 21 ($p \leq 0.05$), 23 ($p \leq 0.01$), and 25 ($p \leq 0.001$). [c] Significant results are shown in bold. [d] Results from Nolan and Farmer (unpublished data); panelists were asked to score for "rancid odor" instead of "off-odor".

Experiment 2: Effect of Low Added Concentrations of Precursors on Odor. The results of paired comparison tests for chicken with precursors added at concentrations close to those occurring naturally are shown in **Table 3**. Significant changes in odor were detected following the addition of ribose, ribose + cysteine, and IMP.

Experiment 3: Effect of Low Added Concentrations of Precursors on Flavor. Further paired comparison tests, designed to investigate the effect on flavor of added food grade precursors (**Table 4**), showed that ribose added at 3−4-fold the natural concentration (100 mg 100 g^{-1}) caused a significant

Table 4. Results of Paired Comparison on the Flavor of Cooked Homogenized Chicken with and without Added Precursors (Experiment 3, Control vs Treated)

precursor	mg added to 100 g of meat[a]	added concn	flavor			
			chicken	meaty	roasted	off
ribose	100	4×	18:12[b]	12:18	**09:21**[c]	**22:08**
glucose	100	2.5×	14:16	14:16	16:14	13:17
IMP	190	2.5×	18:12	14:16	18:12	18:12

[a] Milligrams of compound added to 100 g of minced meat in 10 mL of distilled water. [b] Numbers of panelists selecting control:treated sample as possessing more of this attribute; for 30 panelists. Critical values are 21 ($p \leq 0.05$), 23 ($p \leq 0.01$), and 25 ($p \leq 0.001$). [c] Significant results are shown in bold.

increase in roasted flavor and a decrease in off-flavor as compared with the control.

Experiment 4: Effect of Natural Variation of Ribose and Ribose Phosphate on Flavor of Cooked Chicken Breast. The results for the paired comparison tests of chicken with differing natural levels of ribose indicated that the chicken breast meat with a high concentration of ribose had more roasted chicken flavor than that with low ribose (**Table 5**). However, no difference was observed between chicken breast with high and medium concentrations of ribose (**Table 5**) or for comparisons between leg muscles (**Table 6**).

Experiments 5 and 6: Effect of Selected Precursors on Odor Volatiles. Table 7 shows the principal odors in cooked chicken breast meat with and without added ribose or R5P and those for which a marked change in intensity was observed. Identities are suggested for the compounds responsible.

DISCUSSION

In the experiments described in this paper, precursors of Maillard and related flavor-forming reactions were tested for their contribution to odor and flavor. In experiment 1 (**Table 1**), the quantities of precursors added were based on literature information on the concentrations in chicken meat and/or on previous experiments (Farmer and Nolan, unpublished data). With the benefit of analytical data for the source of chicken used for sensory experiments (*11*, *12*), experiment 2 was designed to screen a range of precursors for their effect on odor, when added at low concentrations similar to those occurring naturally for these (*11*, *12*). The quantity of precursors added was mainly 2−5-fold their natural concentration, which, for many of the precursors, were considerably lower than those used in experiment 1. Subsequent experiments focused on those precursors that gave greatest odor and flavor changes at concentrations close to those found naturally in raw chicken. Simple paired comparison tests using experienced but untrained panelists were used for these latter experiments. The technique was used according to British standard methods (*14*) and was selected to allow comparison of the treatments by panelists more representative of consumers than trained panelists. This Discus-

Table 5. Comparison of Natural Sugar and Sugar Phosphate Concentrations and Sensory Results for Chicken Breast with Low vs High and Medium vs High Concentrations of Ribose

low:high	concentration (low)			concentration (high)			ratio (high/low)			low vs high
chicken	ribose	RP[a]	R[b] + RP	ribose	RP	R + RP	ribose	RP	R + RP	paired comparison
G vs J[c]	10.8	26.0	36.8	29.4	26.3	55.7	2.7	1.0	1.5	02:08
A vs D	7.1	13.0	20.1	22.5	13.5	36.0	3.2	1.0	1.8	05:05
C vs I	12.2	18.8	31.0	31.9	20.3	52.3	2.6	1.1	1.7	02:08
										total: 09:21[d]

low:high	concentration (medium)			concentration (high)			ratio (high/medium)			medium vs high
chicken	ribose	RP[a]	R[b] + RP	ribose	RP	R + RP	ribose	RP	R + RP	paired comparison
L vs K	12.6	6.9	19.5	26.0	14.1	40.1	2.1	2.0	2.1	06:04
H vs E	15.1	28.5	43.6	30.4	20.0	50.4	2.0	0.7	1.2	05:05
F vs B	19.4	3.0	22.4	18.8	19.5	38.3	1.0	6.5	1.7	07:03

[a] RP = ribose phosphate. [b] R = ribose. [c] A–L: Individual chickens with concentrations of ribose classified as low, medium, or high as shown. [d] Numbers of panelists selecting low:high (or medium:high) sample as possessing more roasted chicken flavor. If the assumption is made that the 30 panelists made independent assessments, the critical values are 21 ($p \leq 0.05$), 23 ($p \leq 0.01$), and 25 ($p \leq 0.001$).

Table 6. Comparison of Natural Sugar and Sugar Phosphate Concentrations and Sensory Results for Chicken Leg with Low vs High and Medium vs High Concentrations of Ribose

low:high	concentration (low)			concentration (high)			ratio (high/low)			low vs high
chicken	ribose	RP[a]	R[b] + RP	ribose	RP	R + RP	ribose	RP	R + RP	paired comparison
G vs J[c]	11.6	23.5	35.1	16.6	24.0	40.6	1.4	1.0	1.2	02:03
A vs D	6.4	18.3	24.7	12.4	15.6	28.0	1.9	0.9	1.1	03:02
C vs I	7.1	5.5	12.6	10.7	13.7	24.4	1.5	2.5	1.9	02:03
										total: 07:08[d]

low:high	concentration (medium)			concentration (high)			ratio (high/medium)			medium vs high
chicken	ribose	RP[a]	R[b] + RP	ribose	RP	R + RP	ribose	RP	R + RP	paired comparison
L vs K	11.3	6.9	18.2	13.6	10.8	24.4	1.2	1.6	1.3	04:01
H vs E	11.7	16.4	28.1	20.6	12.7	33.3	1.8	0.8	1.2	02:03
F vs B	11.5	4.9	16.4	11.4	18.9	30.3	1.0	3.9	1.8	02:03

[a] RP = ribose phosphate. [b] R = ribose. [c] A–L: Individual chickens with concentrations of ribose classified as low, medium, or high as shown. [d] Numbers of panelists selecting low:high (or medium:high) sample as possessing more roasted chicken flavor.

Table 7. Principal Odors Detected by GC-O from Cooked Chicken Breast (with and without Sugars)

LRI[a] odor	odor description[b]	suggested identities	method of identification[c]	mean odor scores[d] control	ribose	R-5-P
798	fatty, gas, rotten flesh	hexanal + unknown	MS + LRI + O	0.7	0.5	2.0
817	geranium, metallic, bitter, earthy, pungent	unknown		1.7	2.1	1.4
852	stale beer	unknown		1.8	0.9	1.3
881	chicken, yeast, savory	2-methyl-3-furanthiol	SIM + LRI + O	2.8	3.1	3.5
904	savory, potato	3-methylthiopropanal	MS + LRI + O	0.2	0.8	1.4
907	roasted, savory	2-furanmethanethiol	SIM + LRI + O	0.3	2.1	1.2
967	metallic, geranium, burnt	dimethyltrisulfide + (*E*)-2-heptenal[e]	MS + LRI + O ms + lri + o (*34*)	2.2	2.2	2.7
1076	mushroom	1-nonen-3-one[e]	ms + lri + o (*35*)	1.5	0.8	1.5
1172	chicken, roasted, savory	2-methyl-3-methyldithiofuran[e]	LRI + O	1.4	0.6	2.4
1217	green, metallic	decanal	MS + LRI + O	1.4	0.6	2.4
1762	rotten vegetable	unknown		1.2	0.7	0.4

[a] LRI (CPSIL8CB column). [b] Odors are listed that were detected and described using GC-O by at least two of the four experienced assessors. [c] Identification of compounds. MS, SIM, LRI, O: mass spectrum, SIM ions, LRI, and odor match that of authentic compound analyzed under the same conditions in our laboratory; ms, lri, o: these data match those reported in the literature. [d] Only those odors detected at mean intensity = 2 (or where intensity difference between treatments ≥ 2) are reported. Results are means of duplicate assessments. [e] Compounds in *italics* are tentative identities.

sion will consider the role of each of the potential flavor precursors in turn.

Role of Precursors on the Odor and Flavor of Chicken. *Thiamin.* Thiamin has been shown to be an important precursor of a wide range of sulfur compounds (*3*, *6*, *15*), such as 5-hydroxy-3-mercapto-2-pentanone, which is an intermediate for the formation of many highly odorous thiols, such as 2-methyl-4,5-dihydro-3-furanthiol, 2-methyl-3-furanthiol, and mercaptoketones. In experiment 1, added thiamin gave a pronounced increase in roasted and vegetable soup odors and, with cysteine + ribose, in chicken and savory odors (**Table 2**). However, the concentration of thiamin used for this experiment (340 mg 100 g^{-1}) was extremely high at 1500-fold its natural concentration in chicken breast (**Table 1**). This confirms preliminary results (Nolan and Farmer, unpublished data), which showed that addition of 100 mg 100 g^{-1} (~450-fold) thiamin to raw chicken significantly increased the meaty odor of cooked chicken meat (**Table 3**). In contrast, when added at only twice and 4.5-fold the natural concentration in chicken, 0.50 and 0.90 mg 100 g^{-1}, respectively (experiment 2), thiamin did not cause any significant increase in the odors assessed by paired comparison test (**Table 3**). Thus, while thiamin is, if present at sufficiently high concentrations, a precursor of roasted and meaty odors, the small natural variations (less than 2-fold) observed in commercially produced chicken (*11*) are unlikely to influence flavor.

These findings concur with those from studies on the effect of thiamin on beef odor, which also found that, when added at 4-fold natural concentration, there was no significant effect on odor (*16*). In addition, Grosch et al. (*17*) studied the role of thiamin with cysteine in the formation of 2-methyl-3-furanthiol and suggested that, in meat, the contribution of thiamin to the formation of 2-methyl-3-furanthiol will be very small due to the low natural concentrations of phosphate (0.03 mol L^{-1}). Thiamin breakdown has been shown to be influenced by pH, time (*6*), and water activity (*15*), and changes in these conditions occurring during processing or cooking could change the contribution of thiamin to the flavor of meat.

IMP. No significant effect of IMP on odor was detected by sensory profiling when added to chicken at approximately 5-fold the natural concentration (**Table 2**). Likewise, paired comparison tests showed that IMP, added at once and twice the natural concentration, caused no significant increase in meaty, roasted, or chicken odor (**Table 3**). However, IMP significantly increased the score for off-odor of cooked chicken when added at twice the natural concentration (**Table 3**); from panelists' comments, this descriptor tended to be interpreted as a preference for the control rather than indicative of specific off-odors. Analyses of individual commercial chickens (*11*) showed that concentrations of IMP can vary by up to 2-fold. Thus, it would appear that at concentrations close to those naturally found in raw chicken, differences in IMP concentration have only a small effect on the volatile compounds in cooked chicken.

The role of IMP in chicken flavor formation appears to differ from that found in red meat. Mottram and Madruga (*7*) reported that the addition of IMP (at 10 times the natural concentration) increased the meaty aroma of cooked beef while Farmer et al. (*10*) found that, at 2-fold the reported concentration (170 mg 100 g^{-1}), IMP caused a significant increase in meaty aroma (but not roasted aroma) in pork but not beef. As the concentration of IMP used by these authors in pork is similar to that used for the chicken samples presented in this experiment, it would appear that the impact of IMP may differ between pork and chicken. Analysis of natural concentrations of IMP in the pork and beef used for such experiments and a study of the enzymatic breakdown of such substances may provide further explanation.

Although IMP can be a precursor for 2-methyl-3-furanthiol, mercaptoketones, and other sulfur compounds when reacted with cysteine or H_2S (*18*, *19*), it has been reported (*18*) that, in heated aqueous model systems at pH 5.6, IMP is relatively stable to hydrolysis and therefore does not readily undergo Maillard type reactions with cysteine.

IMP would also be expected to have a role as a flavor enhancer and provider of umami taste (*20*, *21*). However, when IMP + GMP were added at 2.5-fold the natural concentration to minced chicken, no significant effect on flavor was obtained (**Table 4**). However, preliminary studies with chicken burgers suggested that only a doubling (75 mg 100 g^{-1} added) of the natural concentration of IMP may be sufficient to enhance flavor (*22*). Further analyses are needed to determine the conditions that favor the flavor enhancing effects of IMP.

Glucose. Glucose had no significant effect on odor attributes from cooked chicken, whether added at twice the natural concentration and analyzed by paired comparison (**Table 3**) or added at more than four times the natural levels and assessed by profiling (**Table 2**). Neither was flavor affected by the addition of glucose at approximately twice the natural concentration (**Table 4**). The glucose concentration varies between individual commercial chickens by less than many of the sugar-

based precursors, showing ratios of only about two between high and low values (*12*). Thus, it is very unlikely that changes in glucose concentration will cause significant differences in the flavor or odor of cooked chicken meat.

These results agree with those reported previously (*16*) for beef and pork; glucose only affected meaty or roasted aroma at four times the literature concentration for these species; no effect was observed at lower concentrations The low contribution of glucose to flavor-forming reactions is generally attributed to the low reactivity of six-carbon sugars as compared with five-carbon sugars (*23*).

G6P. When added at 300 mg 100 g^{-1}, G6P decreased vegetable/soup odor but did not affect the roasted or chicken aroma of cooked chicken (**Table 2**). Subsequent analyses (*12*) showed that, in chicken meat, the natural concentration of glucose phosphate is, on average, only 16.9 mg 100 g^{-1} and that it varies between individual chickens by up to a factor of 2. No difference was detected between chicken meat with approximately five times (100 mg 100 g^{-1}) this natural concentration added and the control (**Table 3**). Thus, natural variations in G6P appear to have little effect on the odor of chicken.

In contrast, an increase of roasted aroma has been reported with added G6P in beef and pork; however, these additions were at much higher concentrations of 600 and 300 mg 100 g^{-1} (*9*). The natural concentrations of glucose phosphate in beef are also higher than those in chicken, at approximately 80 mg 100 g^{-1} (*12* or *24*−*26*).

The mechanism for the involvement of G6P in flavor forming reactions has not been investigated but might be expected to follow the pathway proposed by van den Ouweland and Peer (*3*) and Mottram and Nobrega (*18*) for R5P. However, G6P can also undergo the pentose phosphate pathway to generate other sugar phosphates.

Ribose. Ribose has long been reported to be an important precursor of meat flavor (*27*). At a relatively high concentration of ribose (150 mg 100 g^{-1}), this precursor significantly increased nutty and woody odors and decreased the rancid and bloody aroma of cooked chicken (**Table 2**). Subsequent analyses (*12*) showed that the natural concentration in chicken breast was only 24.7 mg 100 g^{-1}. Addition at 50 or 100 mg 100 g^{-1} still gave significant increases in chicken, meaty, and roasted aroma together with decreases in off-odor (**Table 3**). Thus, ribose appears to have a significant beneficial effect on the aroma of cooked chicken even when added at only twice or four times the natural concentration.

Similar, but less clear-cut, results have been found for red meats. In cooked beef and pork meat, ribose generally gave increases in both meaty and roasted odors when added at 600 and 1200 mg 100 g^{-1} (*9*), while the effect of adding a lower concentration (127 mg 100 g^{-1}), closer to those later determined in beef, gave a consistent but nonsignificant trend toward increased meaty or roasted odors (*16*). The addition of xylose to ground lamb increased the mild, sweet, meaty aroma and flavors of cooked lamb meat (*8*); the threshold of detection for this effect of xylose was 500 mg 100 g^{-1} meat. Thus, the odor of cooked chicken appears to be affected by lower concentrations of ribose than the red meats.

Further paired comparison tests showed that the same added concentration of ribose (100 mg 100 g^{-1}) also increased the roasted flavor of cooked chicken breast (**Table 4**). A decrease in off-flavor was also observed though; as mentioned previously, this probably indicated a preference for the treated samples rather than detection of a real off-flavor in the control samples.

The addition of precursors into meat provides an indication of their role for odor and flavor, but it is impossible to add them into the same biochemical environment that they would occupy naturally in raw meat. Therefore, an experiment was designed to confirm the above results by comparing the flavor of cooked chicken breast and thighs from chickens with different natural ribose concentrations. Twelve chickens (A−K) were categorized into those with high (six), medium (three), or low (three) ribose concentrations. Paired comparison of low vs high ribose chickens showed that 21 of the 30 panelists assessed the high samples to have the more roasted chicken flavor (**Table 5**). The same effect was not observed for medium vs high ribose chickens. **Table 5** shows that the ratio of ribose between high and low ribose chickens was approximately 2.8, while this ratio was only 1.6 for high and medium chickens. This perhaps suggests that a minimum ribose concentration difference is required to give a perceivable difference in roasted chicken flavor; this concentration difference seems to be about three times, which corresponds to an addition of two times the base concentration. This concurs with the findings of experiment 2 (**Table 3**), which suggested that added ribose at 2−4 times the natural concentration could increase selected odor notes.

Chicken legs (thighs plus drumstick) were also presented to the panelists, but in this case, no significant effect was obtained (**Table 6**). However, each thigh could only be divided into five samples and only 15 panelists could be used for comparison. In addition, the terms low and high were related to the concentration of ribose in chicken breast muscle and not in thighs. **Table 6** shows that the ratios of ribose concentration were generally lower (1.6 for high over low samples), and this ratio may have been insufficient to give a detectable difference in flavor.

Previous studies (*12*) have shown that the natural concentration of ribose can vary both between commercial sources and also between individual chickens. The differences in ribose concentration between a number of the chickens analyzed was in the same order (2−4 times) as the difference observed between high and low ribose chickens in experiment 4. Thus, the variation in ribose concentrations commonly occurring in commercially available chickens appears to be sufficient to give flavor differences in the cooked product and may determine the difference between bland and well-flavored chicken meat.

R5P. When added at 56 and 100 mg 100 g^{-1} [approximately four and six times the natural concentration, respectively (*12*)], R5P did not cause any change in odor (**Table 3**). In experiment 4 (**Table 5**), the concentration of ribose phosphate as well as ribose was recorded for the high, medium, and low ribose chickens. Observation of the results indicates that concentrations of ribose phosphate were practically the same in the comparison of low vs high concentrations of ribose (which gave a sensory difference), while the second group (medium vs high) had a greater difference in ribose phosphate concentrations (but gave no sensory effect). This provides further circumstantial evidence that ribose, rather than R5P, is the more important of these two compounds for flavor and odor formation.

These results differ somewhat from those reported in model systems and beef. Mottram and Nobrega (*18*) reported a higher reactivity of R5P than ribose in producing larger quantities of volatile compounds (e.g., mercaptoketones and their disulfides) using model systems containing cysteine with ribose or R5P. R5P is believed to be a precursor of furan and thiophenethiols via the dephosphorylation and dehydration of ribose phosphate, which forms the intermediate 4-hydroxy-5-methyl-3(2H)-furanone, which readily reacts with hydrogen sulfide (*3*). When

added to beef at approximately 30-fold the natural concentration, R5P causes significant increases in the concentrations of furanthiols and disulfides. Sensory profiling studies on the aroma of the cooked beef (*10*) showed that both ribose and R5P increased the roasted aroma.

Cysteine. While cysteine is one of the more difficult amino acids to analyze, results (*11*) suggest that the concentration of free cysteine is extremely low (<0.1 mg 100 g^{-1}). The addition of substantially greater than this concentration (120 mg 100 g^{-1}, **Table 2**) gave increases in bloody and sour notes, while much lower concentrations (0.7 mg 100 g^{-1}, **Table 3**) had no effect on sensory attributes. In contrast, when combined with ribose, higher scores for chicken and salty odors were obtained (**Table 2**) together with a more roasty odor (**Table 3**). These results suggested that the limiting factor for increasing these odors is ribose rather than cysteine. Thus, although the role of cysteine is crucial for the formation of S-containing volatile compounds, the results presented herein suggest that its natural concentration was sufficient for generating S-containing volatile compounds.

The very low apparent concentrations of free cysteine present in raw meat as compared with reducing sugars suggest that this amino acid is not a prerequisite of the Maillard reaction with ribose to yield important flavor compounds. It would seem probable that other, more abundant, amino acids fulfill this role while cysteine, in its protein form, serves primarily as a source of H_2S. Early work by Mecchi et al. (*28*) proposed that protein cysteine is the major source of H_2S in cooked meat.

Effects of Selected Added Precursors on Odor Volatiles. To determine whether the sensory results reported above could be explained by the volatile compounds formed, studies were also conducted, using GC-O, to assess key odor volatiles. The effect of added ribose and R5P at four and six times, respectively, the natural concentration in raw meat on the odor volatiles generated in cooked chicken is shown in **Table 7**. A static headspace collection method was used for GC-O as it represented more closely the quantities of aroma perceived by panelists. The aim of this method is to present the odors at a concentration close to that perceived in real life, so that any changes in volatile concentrations have a similar impact as they would in a real food. Dynamic headspace concentration was used for GC-MS identification.

The principal odors detected by GC-O (**Table 7**) included roasted, savory, chicken, and metallic odors. Some showed an increase in intensity on addition of ribose or R5P, especially savory, potato (904) and roasted, savory (907). These odors were caused by 3-methylthiopropanal and 2-furanmethanethiol. Roasted, meaty odors at 881 (2-methyl-3-furanthiol) and 1172 (probably 2-methyl-3-methyldithiofuran) showed only small increases in odor intensity. However, these odors were perceived as sufficiently intense that intensity differences might not have been detected. A preliminary study to quantify 2-methyl-3-furanthiol and 2-furanmethanethiol by SIM (with identification confirmed using LRI, three ions, and their ratios) gave peak areas in the ratio 1:3.5:2 (2-methyl-3-furanthiol, ion 114) and 1:4:2 (2-furanmethanethiol, ion 81) for control chicken and that with added ribose and R5P, respectively. These data suggest that the actual quantities of both compounds were affected by the added sugars.

Several odors apparently reduced in intensity on addition of ribose. Odors of stale beer (852; unknown), mushroom (1076, probably 1-nonen-3-one), rotten vegetable (1762, unknown) decreased in intensity; this last was also affected by R5P. Further work is needed to determine if this is an indication of the phenomenon observed previously (*9*) that addition of sugars can reduce quantities of aliphatic compounds formed.

Thus, the increases observed in sensory trials of roasted, meaty, and chicken odor and flavor, following small additions of ribose (but not R5P) are probably due to increases in the quantities of 2-furanmethanethiol and, probably, 2-methyl-3-furanthiol and 3-methylthiopropanal. Other compounds may also be involved in changing the overall balance of odor and flavor.

LITERATURE CITED

(1) Mottram, D. S. Meat. In *Volatile Compounds in Foods and Beverages*; Maarse, H., Ed.; Marcel Dekker: New York, NY, 1991; pp 107−177.

(2) Farmer, L. J. Poultry meat flavour. In *Poultry Meat Science*; Richardson, R. I., Mead, G. C., Eds.; CABI Publishing: Wallingford, United Kingdom, 1999; pp 127−158.

(3) van den Ouweland, G. A. M.; Peer, H. G. Components contributing to beef flavor. Volatile compounds produced by the reaction of 4-hydroxy-5-methyl-3(2H)-furanone and its thio analogue with hydrogen sulfide. *J. Agric. Food Chem.* **1975**, *23*, 501−505.

(4) Hofmann, T.; Schieberle, P. Quantitative model studies on the effectiveness of different precursor systems in the formation of the intense food odorants 2-furfurylthiol and 2-methyl-3-furanthiol. *J. Agric. Food Chem.* **1998**, *46*, 235−241.

(5) Madruga, M. S.; Mottram, D. S. The effect of pH on the formation of Maillard-derived aroma volatiles using a cooked meat system. *J. Sci. Food Agric.* **1995**, *68*, 305−310.

(6) Güntert, M.; Bruning, J.; Emberger, R.; Kopsel, M.; Kuhn, W.; Thielmann, T.; Werkhoff, P. Thermally degraded thiamin. A potent source of interesting flavour compounds. In *Flavour Precursors. Thermal and Enzymatic Conversions*; Teranishi, R., Takeoka, G. R., Güntert, M., Eds.; American Chemical Society: Washington, DC, 1992; pp 140−163.

(7) Mottram, D. S.; Madruga, M. S. The role of inosine monophosphate as a precursor of meat aroma. In *Trends in Flavour Research*; Maarse, H., van der Heij, D. G., Eds.; Elsevier Science: Amsterdam, The Netherlands, 1994; pp 339−344.

(8) Hudson, J. E.; Loxley, R. A. The effect of pentose sugars on the aroma and flavour of mutton. *Food Technol. Aust.* **1983**, *35* (4), 174−175.

(9) Farmer, L. J.; Hagan, T. D. J.; Paraskevas, O. A comparison of three sugars and inosine monophosphate as precursors of meat aroma. In *Flavour Science: Recent Developments*; Taylor, A. J., Mottram, D. S., Eds.; Cambridge, United Kingdom, 1996; pp 225−230.

(10) Farmer, L. J.; Hagan, T. D. J.; Paraskevas, O. Role of selected precursors in meat flavour formation. In *Quality Attributes of Muscle Foods*; Xiong, Y. L., Ho, C.-T., Shahidi, F., Eds.; Plenum Publ. Corp.: New York, NY, 1998; pp 159−172.

(11) Aliani, M.; Farmer, L. J. Precursors of chicken flavor. I. Determination of some flavor precursors in chicken muscle. *J. Agric. Food Chem.* **2005**, *15*, 6078−6083.

(12) Aliani, M.; Farmer, L. J. A postcolumn derivatization method for determination of reducing and phosphorylated sugars in chicken by high performance liquid chromatography. *J. Agric. Food Chem.* **2002**, *50*, 2760−2766.

(13) Chevance, F. F. V.; Farmer, L. J. Identification of major volatile odor compounds in frankfurters. *J. Agric. Food Chem.* **1999**, *47*, 5151−5160.

(14) BS 5929: Part 2. Paired comparison test. British standard methods for sensory analysis of food, 1982.

(15) Hartman, G. J.; Carlin, J. T.; Scheide, J. D.; Ho, C.-T. Volatile products formed from the thermal degradation of thiamin at high and low moisture levels. *J. Agric. Food Chem.* **1984**, *32*, 1015−1018.

(16) Farmer, L. J.; Hagan, T. D. J. Precursors of flavour in cooked beef. In *Proceedings of ICOMST*; Università di Parma: Rome, Italy, 2002; pp 322−323.

(17) Grosch, W.; Zeiler-Hilgart, G.; Cerny, C.; Guth, H. Studies on the formation of odorants contributing to meat flavours. In *Progress in Flavour Precursor Studies*; Schreier, P., Winterhalter, P., Eds.; Allured Publishing: Carol Stream, IL, 1993; pp 329−342.

(18) Mottram, D. S.; Nobrega, I. C. C. Formation of volatile sulphur compounds in reaction mixtures containing cysteine and three different ribose compounds. In *Food Flavors: Formation, Analysis and Packaging Influences: Proceedings of the 9th International Flavor Conference*; Contis, E. T., Spanier, A. M., Shahidi, F., Eds.; Elsiever: Amsterdam, The Netherlands, 1998; pp 483−492.

(19) Zhang, Y.; Ho, C. T. Formation of meatlike aroma compounds from thermal reaction inosine 5′-monophosphate with cysteine and glutathione. *J. Agric. Food Chem.* **1991**, *39*, 1145−1148.

(20) Sugita, Y. H. In *Developments in Food Flavours*; Birch, G. G., Lindley, M. G., Eds.; Elsevier Applied Science: London, United Kingdom, 1986; pp 63−79.

(21) Maga, J. A. Umami flavor in meat. In *Flavor of Meat and Meat Products*; Shahidi, F., Ed.; Chapman and Hall: Glasgow, 1994; pp 98−115.

(22) Aliani, M. *Investigation of the Key Flavour Precursors in Chicken Meat*; Queen's University Belfast: Belfast, 2002.

(23) Spark, A. A. Role of amino acids in nonenzymatic browning. *J. Sci. Food Agric.* **1969**, *20*, 308−316.

(24) Lawrie, R. A. *Meat Science*, 3th ed.; Pergamon Press: Oxford, United Kingdom, 1979; p 267.

(25) Pearson, A. M.; Young, R. B. Postmortem changes during conversion of muscle to meat. In *Muscle and Meat Biochemistry*; Academic Press: New York, 1989.

(26) Paraskevas, O. Studies on carbohydrates in meat in relation to colour and flavour. M.Philos., The Queen's University of Belfast, Belfast, 1997.

(27) MacLeod, G. M.; Seyyedain-Ardebili, M. Natural and simulated meat flavors (with particular reference to beef). *CRC Crit. Rev. Food Sci. Nutr.* **1981**, *14*, 309−437.

(28) Mecchi, E. P.; Pippen, E. L.; Line-Weaver, H. Origin of hydrogen sulfide in heated chicken muscle. *J. Food Sci.* **1964**, *29*, 393−399.

(29) Fujimura, S.; Kawano, S.; Koga, H.; Takeda, H.; Kadowaki, M.; Ishibashi, T. Identification of taste-active components in the chicken meat extract by omission test. Involvement of glutamic acid, IMP and potassium ion. *Anim. Sci. Technol. (Jpn.)* **1995**, *66*, 43−51.

(30) Lilyblade, A. L.; Peterson, D. W. Inositol and free sugars in chicken muscle post-mortem. *J. Food Sci.* **1962**, *27*, 245−249.

(31) Schliemann, J.; Wolm, G.; Schrodter, R.; Ruttloff, H. Huhnerfleischaroma-Bildung, zusammensetzung und gewinnung 1.Mitt. aromavorlaufer. *Nahrung* **1987**, *31* (1), 47−56.

(32) Abdulrahman, S. A.; Abdelbary, A. D. Effects of cooking methods on thiamin and riboflavin contents of chicken meat. *Food Chem.* **1993**, *48* (1), 69−74.

(33) Leonhardt, M.; Wenk, C. Animal species and muscle related differences in thiamine and riboflavin contents of Swiss meat. *Food Chem.* **1997**, *59* (3), 449−452.

(34) Ullrich, F.; Grosch, W. Identification of the most intense volatile flavour compounds formed during autoxidation of linoleic acid. *Z. Lebensm. Unters. Forsch.* **1987**, *184*, 277−282.

(35) Roberts, D. D.; Acree, T. E. Effects of heating and cream addition on fresh raspberry aroma using a retronasal aroma simulator and gas chromatography olfactometry. *J. Agric. Food Chem.* **1996**, *44*, 3919−3925.

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