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饲料的分子结构与动物营养物质利用率和有效性的关系——一种新方法

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摘要

新的研究思路、研究方法和生物分析技术的创造和发展对动物科学（包括饲料和营养科学）的进步是必不可少的。本文介绍了以同步加速器为基础的先进生物分析技术作为一项崭新的研究工具，在研究由多种处理（如基因修饰、基因沉默、饲料的热加工处理、生物燃料加工等）诱导的饲料分子结构变化与动物消化吸收饲料营养物质的关系方面的潜在应用。以同步辐射为基础的先进技术 [如同步辐射红外显微光谱技术（synchrotron radiation infrared microspectroscopy，SR-IMS）和同步辐射 X 射线技术] 作为一种快速、无损的生物分析技术被开发利用。与传统的湿化学法不同，同步辐射技术不会破坏饲料内在的分子结构。尖端和先进的同步加速器光源（是日光的上百万倍）能够以超高分辨率在细胞和分子水平上探测生物组织的内在结构。总的来说，最近开发的基于同步辐射的生物分析技术结合常用的研究技术将带来动物饲料和营养研究的巨大进步。

1. 引言

动物科学包括饲料和营养科学，它的发展离不开新的研究思路、研究方法和生物分析技术的创造和发展[1]。常用的传统的湿化学法经常用于营养物质的分析和饲料评价。然而，湿化学法经常破坏饲料的内在分子结构，因为湿化学法在饲料消化制备和分析处理的过程中应用了大量刺激性的化学试剂[1–3]。这些化学物质经常破坏或改变饲料的固有或原来的分子结构，并且常常产生影响饲料和营养评价的次生物质[1–2]。

最近开发的先进的同步辐射红外显微光谱技术（SR-IMS）是一种快速、无损、直接的生物分析技术[4–7]。这种尖端的生物分析技术的辐射光亮度强（是日光的上百万倍）、发散性低、有效源尺寸小[5,6]。它能够以超高分辨率探测生物组织的分子结构[4,8–12]。使用以同步加速为基础的生物分析技术使同时获取组织结构、组织营养、组织化学和组织环境等多种类型的信息（图1）成为可能[2,13,14]。

目前，SR-IMS在研究饲料分子结构与分子营养或传统的动物营养之间的相关关系方面的应用较少。同样的，利用先进的以同步加速为基础的生物分析技术探测动物饲料细胞和亚细胞水平上的内在结构组成的应用也

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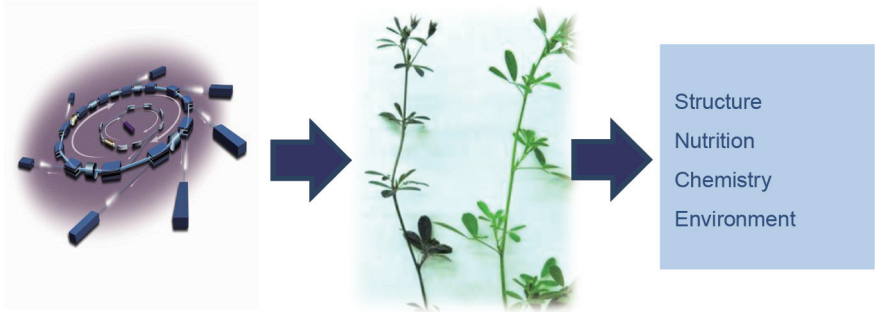

图1. 以先进的同步辐射技术为基础的生物分析技术能够同时提供四种类型的信息，包括组织结构、组织营养、组织化学、组织环境。

极少，而饲料的结构组成与动物营养的传递直接相关[15,16]。伴随其他因素（如营养素），饲料内在的分子结构和组成会影响饲料品质、营养价值和生物学功能及其在动物体内的发酵、降解动力学及消化过程[12,17]。饲料的分子构造和组成强烈地影响蛋白质在小肠内的吸收率，并且影响蛋白质与动物消化道内源消化酶的接触[18,19]。蛋白质与消化酶接触的减少会导致蛋白质消化和吸收率下降，从而降低其对动物的营养价值[20–22]。

本文的目的是介绍我们新的研究思路和生物分析技术（即以同步辐射为基础的生物分析技术），并以此作为动物科学领域研究饲料分子营养和分子结构之间的定量交互关系的一种新方法。

本文包括以下几部分：第2部分介绍了以同步加速器为基础的生物分析技术的概念、同步加速装置的主要组成部件及一些同步辐射分子光谱技术；第3部分基于同步辐射研究项目介绍了这项技术的应用；第4部分是本文的结论。

2. 基于新的以同步加速为基础的生物分析技术的营养和饲料研究项目

2.1. 同步加速辐射设备的概念

什么是同步加速器？简单来说，同步加速器是一个把电子变成光的巨大粒子加速器 [4–6]。同步辐射装置包括多个组成部件，如电子枪、直线加速器、增能环、储存环、很多光束线（如红外线、软X射线和硬X射线）和终端实验站[4–6]。一个中等规模的同步辐射装置大约是一个足球场的大小。位于加拿大萨斯喀彻温省的萨斯喀彻温大学里的加拿大国家同步辐射设施——加拿大光源(CLS)是中等规模同步辐射装置的一个例子。然而，很多同步辐射装置规模较大，如美国伊利诺斯州芝加哥的先进的光子源、美国纽约州厄普顿的国家同步辐射光源II及日本兵库县的播磨科学花园城的SPring-8。同步辐射装置的规模大小部分取决于同步加速器的目标能级（从0.8 GeV到8.0 GeV）。

2.2. 利用同步辐射装置研究饲料分子结构

同步辐射光亮度极高，它是一种全波段光子束和电磁辐射源。加速器使电子加速获得高能，同步辐射装置中的弯转磁铁和波荡器使高能电子束转换为光子束，这个光子束被称为同步加速器辐射光。科学家们在同步辐射加速器光束线实验站通过分析同步辐射光谱来研究饲料分子结构[1,5,6,23,24]。应用这一技术的唯一缺点是需要使用同步辐射装置，而它的造价高达数百万美元。

2.3. 利用以尖端同步加速为基础的生物分析技术研究植物酰胺

植物性饲料、种子、绿色饲草及青贮饲料的蛋白质具有独特的分子化学组成和分子结构，所以每一种植物性饲料蛋白的分子光谱是唯一的。植物性饲料蛋白的光谱在振动的中红外光谱区，有两个重要且显著的特征：蛋白质氨基I光谱，峰值在1600~1700 cm^{-1}；蛋白质氨基II光谱，峰值在1500~1560 cm^{-1}。这两个特有的光谱峰是由蛋白质主链的振动（即拉伸和弯曲）[25–28]产生。植物性饲料的蛋白质氨基I光谱（而不是II光谱）经常用于蛋白质α螺旋、β折叠、无规则卷曲、β转角等结构的分析[29,30]。

2.4. 利用以尖端的同步加速为基础的生物分析技术进行多变量分子光谱分析

多变量技术或者方法可以用来分析植物性饲料的分子光谱，发现饲料分子结构的差别。凝聚层次聚类分析分子光谱和主成分分析是多变量分析方法中的两种。这些多变量分析法不需要知道光谱分布，其目的仅仅是区分和定性地分离出影响分子结构和诱导分子结构

变化的处理，因为结构的变化可能影响动物对饲料养分的吸收[30–33]。

3. 基于同步加速的生物分析技术在饲料和分子营养中的研究和应用

3.1. 应用 I：动物饲料的分子化学成像

利用同步辐射为基础的生物分析技术的首次应用包括动物饲料的分子化学成像[31]。这种应用的例子包括小麦[4]、先锋玉米[34]和高粱[13]的分子化学成像。这些研究是我们团队在美国布鲁克海文国家实验室（NSLS-BNL，美国能源部）的同步加速光源或者加拿大CLS进行的。这项研究是为了探讨加工处理对生长在加拿大西部地区黄色种皮的甘蓝型油菜籽子叶组织的影响[14]。利用饲料分子成像技术，我们也可以观测到受霜冻或冷冻损坏的谷物种子和正常种子（如小麦）之间的差异。

3.2. 应用 II：由基因转化和基因修饰诱导引起的植物源饲料分子结构变化的检测

第二项应用是利用同步辐射为基础的生物分析技术检测那些由基因转化、基因插入或者基因沉默诱导的蛋白质组成或构造的外部结构变化。这项研究是由我们团队在NSLS-BNL和美国劳伦斯伯克利国家实验室（ALS-LBNL）的先进光源实验室进行的。我们的团队利用尖端的同步加速技术开展了比较和区分普通苜蓿蛋白（即无外源基因插入）和转基因苜蓿植物组织（亚细胞水平上插入外源的玉米*Lc*调节基因）的分子结构差异的研究。另外，我们还利用Gauss-Lorentz方法建立多组分峰模型量化了蛋白质生物聚合物中的构型[35–38]。

目前，我们的团队正在利用以同步加速为基础的生物分析技术研究插入双基因和两个外源基因对苜蓿分子结构的影响[39,40]，以及基因沉默对苜蓿结构变化的影响[41]。所有这些结构研究都同养分的传递研究相关联。我们的研究结果表明，对于只放牧在苜蓿牧场、干物质摄入量为17 kg、体重为650 kg的奶牛，单个*Lc*基因转化可使每头奶牛每天多产2 kg奶[36–38]。

3.3. 应用 III：在分子水平上检测热诱导引起的饲料结构变化

第三项应用是利用以同步辐射为基础的生物分析技术检测热诱导蛋白的结构和亚组分特征对反刍动物瘤胃蛋白质降解以及肠道消化的影响[9,10,42,43]。我们的团队利用先进的同步加速技术作为一种新的工具和方法进行了几项研究。这些研究表明饲料组织的内部结构受多种处理的影响，并且定量了蛋白质结构与营养之间的互作关系[9,10,19,41–45]。作为一种先进的工具，同步加速技术不仅使得研究蛋白质生物聚合物的内部分子结构成为可能，也使得研究碳水化合物的分子结构[17]以及脂质生物聚合物的结构组成成为可能。在反刍动物饲粮配方中，总代谢蛋白质和降解蛋白质平衡是最为重要的两个指标。有了这个新的工具，这两个指标有可能通过建立的基于饲料分子结构特征的方程来预测，而不用通过费时且昂贵的奶牛代谢试验（图2）。

3.4. 应用 IV：生物乙醇加工对饲料内部分子结构影响的研究

第四项应用是利用以同步加速为基础的生物分析技术检测由生物能源/生物燃料加工引起的分子结构变化。

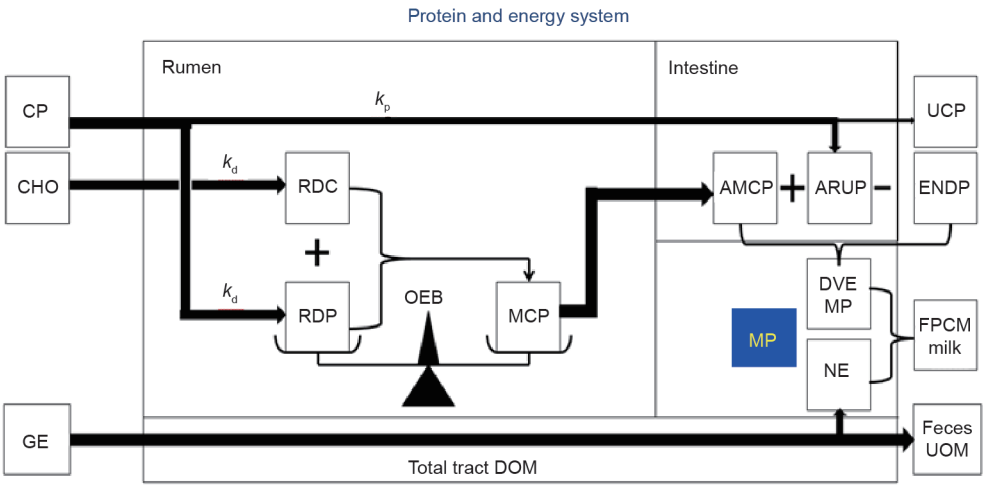

图2. 测定饲料或日粮中的可代谢蛋白质的费时且昂贵的试验过程。

我们团队还研究了蛋白质生物聚合物同其在动物体内代谢特征之间的互作关系[16]。这项研究是为了揭示生物能源生产过程中产生的蛋白质的分子组成和构象，并区分起初的原料与各种副产品之间的差异[16]。我们的团队目前正在利用这种先进技术来发现能源加工过程中新的副产物[如藻粕（carinata meal）]，并将其与传统的生物油加工副产物相比较（如双低菜籽粕），以作为奶牛和肉牛的新的饲料原料来源[45,47]。

4. 总结与启示

总而言之（图3），正如本文所述，我们的研究团队在分子基础上提供了一个先进的饲料和营养研究的新概念。这些研究表明同步加速技术在以超高空间分辨率展示不同处理和加工后家畜饲料内部分子构象变化的潜力。这种尖端的技术可以用来揭示反刍动物和单胃动物的养分吸收与分子结构变化之间的互作关系。

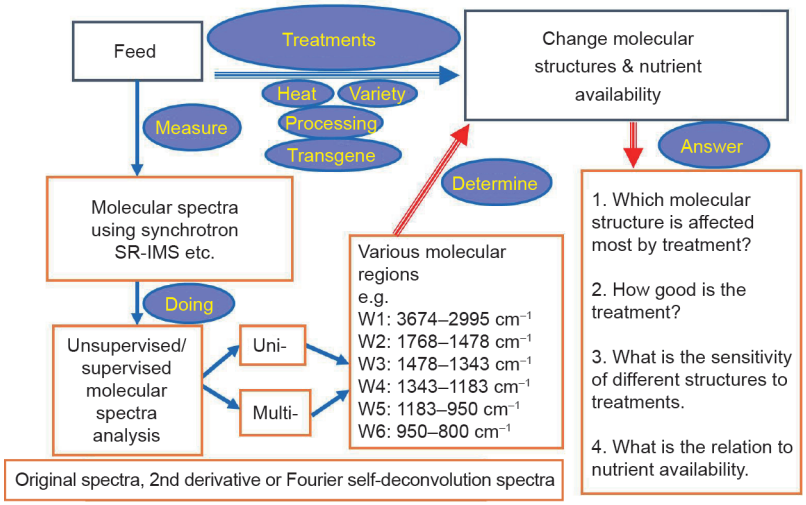

图3. 以同步辐射为基础的分子光谱技术方法的总结和应用。

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Compliance with ethics guidelines

Peiqiang Yu and Luciana L. Prates declare that they have no conflict of interest or financial conflicts to disclose.

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