

引用格式: LIU Wenbo, YAO Mingfei, CHEN Jiayu, et al. Comparison of Near-Infrared Diffuse Reflectance Spectral Responses and Skin Microcirculatory Changes Under Glucose and Whey Protein Loading[J]. Acta Photonica Sinica, 2027, 56(1): 0130001
刘文博, 姚明飞, 陈嘉宇, 等. 葡萄糖和乳清蛋白负荷下的近红外漫反射光谱响应及其皮肤微循环变化的比较[J]. 光子学报, 2027, 56(1): 0130001

葡萄糖和乳清蛋白负荷下的近红外漫反射光谱响应及其皮肤微循环变化的比较

刘文博¹, 姚明飞², 陈嘉宇¹, 陈宇恒¹, 孙迪², 刘瑾¹, 韩同帅¹

(1 天津大学 精密测试技术及仪器全国重点实验室, 天津 300072)

(2 先阳科技(天津)有限公司, 天津 300192)

摘 要:在基于近红外漫反射光谱法的葡萄糖负荷研究中, 进食后机体会出现血糖升高、胰岛素分泌增加、血流灌注改变等复杂生理反应, 使得光谱信号来源具有多源性。其中, 胰岛素水平的快速变化是诱发血流灌注等反应的重要因素之一。为考察胰岛素分泌等代谢变化对光谱测量信号的影响, 并尽量避免血糖波动干扰, 本文设计了乳清蛋白负荷实验, 并与葡萄糖负荷实验进行对比。实验中, 6 名健康受试者分别摄入 100 mL 含 20 g 葡萄糖或乳清蛋白的水溶液, 其中 2 名受试者同步监测血清胰岛素浓度及真皮浅层微循环血流水平。结果显示, 摄入后 30 min, 两种负荷条件下胰岛素浓度均明显升高, 但激光多普勒血流仪检测到的真皮浅层微循环血流信号未发生显著变化。两种负荷条件下采集的 1050 - 1609 nm 波段的光谱信号表现出明显差异。葡萄糖负荷后, 光谱信号均出现显著变化, 其中血糖信号最灵敏的 1550 nm 处漫反射差分吸光度与血糖变化呈较好相关性; 而乳清蛋白负荷下光谱信号未发生显著变化。综上, 在本实验条件下, 乳清蛋白负荷诱导的胰岛素分泌等代谢变化对真皮浅层微循环血流影响幅度较小, 且对采集的 1050 - 1609 nm 波段的光谱信号影响也较小。

关键词:无创血糖测量; 激光多普勒血流测量; 近红外漫反射光谱法; 胰岛素; 乳清蛋白

中图分类号: O433.4

文献标识码: A

doi: 10.3788/gzxb20275601.0130001

0 引言

进食后, 机体会触发一系列复杂的生理反应, 这些变化可能导致无创血糖测量中的光谱信号来源呈现多源性。除血糖自身贡献外, 由生理扰动诱发的光谱响应也可能混入测量信号中, 从而对血糖检测准确性构成干扰。因此, 识别这些干扰来源, 对于提升无创血糖测量的可靠性具有重要意义。

在众多可能的影响因素中, 胰岛素分泌相关的代谢响应及其引起的微循环变化可能是潜在影响因素之一。以口服葡萄糖耐量试验(oral glucose tolerance test, OGTT)为例, 血糖升高不仅可直接改变组织光学参数, 如吸收系数、散射系数及折射率等^[1-2], 还可迅速刺激胰岛素分泌, 以促进细胞对葡萄糖的摄取和利用^[3-5]。同时, 胰岛素作为连接餐后代谢反应与血管功能变化的重要体液调节激素, 可作用于血管内皮细胞, 激活内皮型一氧化氮合酶, 促进一氧化氮生成, 进而诱导微血管舒张及毛细血管募集, 并伴随一定热效应, 最终增加局部血流灌注以及血管与间质间的物质交换面积^[6]。

本课题组 Han 等^[7]在人体实验中发现, 在 15 例受试者开展的 17 次 OGTT 中, 1550 nm 单波长处光信号与血糖变化之间的平均相关系数约为 0.84。然而, 不同组次 OGTT 实验之间的相关性存在明显差异, 最低

基金项目:国家自然科学基金(81971657)

第一作者:刘文博(1999—), 男, 博士研究生, 主要研究方向为无创血糖测量。Email: wenbo_liu@tju.edu.cn

通讯作者:刘瑾(1979—), 女, 副教授, 博士, 主要研究方向为光谱检测与光谱分析。Email: liu_jin@tju.edu.cn; 韩同帅(1991—), 男, 助理研究员, 博士, 主要研究方向为无创血糖测量。Email: hts2014@tju.edu.cn

收稿日期: 2026-07-02; **录用日期:** 2026-08-05

<http://www.photon.ac.cn>

相关系数仅为 0.5,最高可达到 0.95。随后,课题组采用相同检测系统开展了进一步的人体实验研究(如 Ge 等^[8]报道了多组实验结果),并对部分受试者进行了多次重复测量。结果显示,同一受试者不同次 OGTT 实验获得的相关系数同样存在明显波动。上述现象表明,除血糖变化本身、个体差异外,可能还存在其他未知的生理因素影响光谱测量信号。然而,在现有实验条件下,这些因素的影响尚未超过血糖信号的主导作用,其主要表现为降低单波长血糖测量结果的精密度。因此,本文首先针对胰岛素分泌相关生理响应可能引起的影响展开探究,设计了乳清蛋白负荷诱导胰岛素分泌增强实验,以探究由胰岛素相关生理响应引起的微循环血流变化及其在光谱信号中的表现。需要指出的是,乳清蛋白负荷除可促进胰岛素分泌外,还可能同时诱导其他生理变化,例如血浆氨基酸水平升高以及肠促胰素分泌变化等^[9]。因此,若乳清蛋白负荷后微循环血流参数及光谱信号均未出现显著变化,则可初步排除胰岛素作为主要影响因素的可能性;若观察到显著变化,则该变化还不能直接归因于胰岛素作用,仍需结合其他实验进一步明确其具体生理来源。

为避免血糖波动带来的混淆效应,本研究设计了乳清蛋白负荷实验。乳清蛋白具有较强的促胰岛素分泌作用,摄入后可升高血浆氨基酸水平,并通过直接刺激胰岛 β 细胞及促进肠促胰素释放等途径增强胰岛素分泌^[9]。研究表明,在健康受试者中,1 g 葡萄糖可引起约 4.7 pM 的胰岛素升高,而 1 g 乳清蛋白可引起约 4.8 pM 的胰岛素升高,二者在同等质量下对胰岛素分泌的刺激效应相近^[10]。因此,相较于葡萄糖负荷,乳清蛋白负荷更适合用于构建一种以胰岛素升高为主要特征、且在空腹状态下几乎不影响血糖水平的餐后模型,从而有助于观察胰岛素相关的皮肤微循环变化。基于这一特点,乳清蛋白负荷实验可为区分胰岛素效应与葡萄糖效应提供更清晰的实验框架。

激光多普勒血流仪(laser Doppler flowmetry, LDF)是评估皮肤微循环血流变化的常用方法,可用于观察胰岛素刺激下的皮肤血流反应^[11]。Meijer 等^[12]招募 18 名健康志愿者,对比了在生理盐水输注和高胰岛素-正常血糖钳夹条件下,胰岛素对皮肤及骨骼肌微循环功能的影响。结果显示,在皮肤微循环中,高胰岛素状态并未改变连续灌注毛细血管数量(生理盐水输注:40.8 $\pm$ 8.3 n/mm²;高胰岛素-正常血糖钳夹:40.3 $\pm$ 8.6 n/mm²)。在骨骼肌微循环中,高胰岛素状态显著增加了微血管血容量,由 7.00(生理盐水输注状态)升高至 10.06(高胰岛素-正常血糖钳夹状态)。Hohberg 等^[13]观察了 15 名 2 型糖尿病患者注射人胰岛素后皮肤微血管血流的变化,结果显示,人胰岛素组皮肤微血管血流由 27.9 AU 轻度升至 34.4 AU。上述研究表明,胰岛素刺激下皮肤微循环血流响应相对微弱,而骨骼肌等代谢活跃的深层组织的血流反应则更为显著。考虑到近红外光谱测量具有较高的响应灵敏度,即使胰岛素仅使得皮肤微循环发生微小变化,理论上仍可能对光谱信号产生可测影响,因此有必要在实际测量条件下考察其对光谱响应的实际贡献。

此外,皮肤微循环血流的变化还可能改变毛细血管灌注状态,进而影响组织间质液的含量,最终对血糖测量产生潜在干扰^[14-15]。而 LDF 主要反映采样体积内运动红细胞浓度和平均运动速度的综合变化,能够评价局部血流灌注水平^[16],但难以反映血管外间质液含量的变化。相比之下,1050 - 1609 nm 宽波段近红外漫反射光谱对组织吸收和散射变化均具有响应,可同时反映血液相关变化及组织水分变化,因此可作为 LDF 测量的重要补充。此外,本文采用的近红外光谱测量与 LDF 方法具有相近的组织探测深度,均主要反映浅层真皮区域的微循环状态,有助于进一步分析胰岛素分泌增强后该区域血流灌注变化对光谱信号的潜在影响。

综上所述,本研究设计并开展了葡萄糖负荷与乳清蛋白负荷对比实验,同步测量血清胰岛素浓度、近红外漫反射光谱及激光多普勒血流信号,重点考察胰岛素升高期对皮肤微循环及其光学信号的影响。该研究可为识别无创血糖测量中的潜在生理干扰因素提供实验依据与参考。

1 方法

1.1 近红外漫反射光谱法

皮肤组织中近红外漫反射光谱法的简化模型如图 1 所示^[17]。入射近红外光在表皮、真皮及皮下组织三层结构中发生吸收与散射,并在多次散射后由皮肤表面近光源处返回检测端。组织葡萄糖浓度变化会对近红外光的吸收及散射特性产生一定影响。因此,可通过分析近皮肤表面漫反射光谱吸光度的变化,间接表征局部分析物浓度的改变。在给定光源-检测器距离(Source-Detector Separation, SDS)条件下,检测到的

漫反射光吸光度可定义为:

$$A(\lambda, \rho) = -\ln \left(\frac{I(\lambda, \rho)}{I_0(\lambda)} \right) \quad (1) \text{ 其中 } \lambda \text{ 为检测使用的波长, } \rho \text{ 为 SDS, } I_0$$

为入射光强, I 为出射光强。本研究采用两个不同 SDS 上的吸光度差分策略, 可以有效消除部分活体测量过程中系统漂移和人体背景漂移等共模干扰^[7]。差分吸光度定义如下:

$$A_D(\lambda) = A(\lambda, \rho_2) - A(\lambda, \rho_1) = \ln \frac{I(\lambda, \rho_1)}{I(\lambda, \rho_2)} \quad (2)$$

其中 ρ_1 与 ρ_2 分别为 I_1 与 I_2 处的 SDS。

三层皮肤组织在 1000 - 1700 nm 波长范围内的平均穿透深度及血糖差分信号灵敏度如图 2 所示^[18-19]。从平均穿透深度来看, 多数波长的平均穿透深度位于 1 - 2 mm 范围内, 主要聚焦于皮肤真皮区域^[20]。不同波段对组织主要成分的敏感性存在差异: 1000 - 1100 nm 附近波长对血红蛋白吸收较为敏感; 1200 nm 附近波长对脂质吸收较为敏感; 1300 nm 附近波长受水吸收影响较小, 可作为反映组织散射变化的参考波段; 而 1400 - 1650 nm 附近波长对水和葡萄糖吸收较为敏感^[21-22]。综合考虑组织穿透深度、主要吸收成分及血糖差分信号灵敏度, 本文选取 1050、1219、1314、1409、1550 和 1609 nm 作为皮肤近红外探测波长。

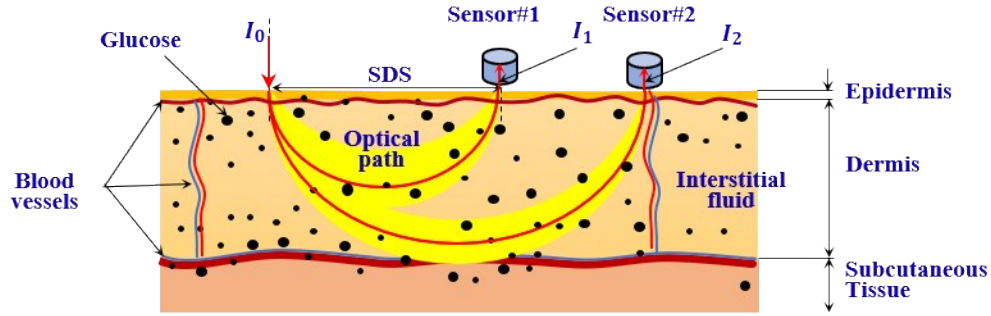


图1 皮肤组织近红外漫反射光谱法简化模型

Fig.1 Simplified model of NIR diffuse reflectance spectroscopy in skin tissue

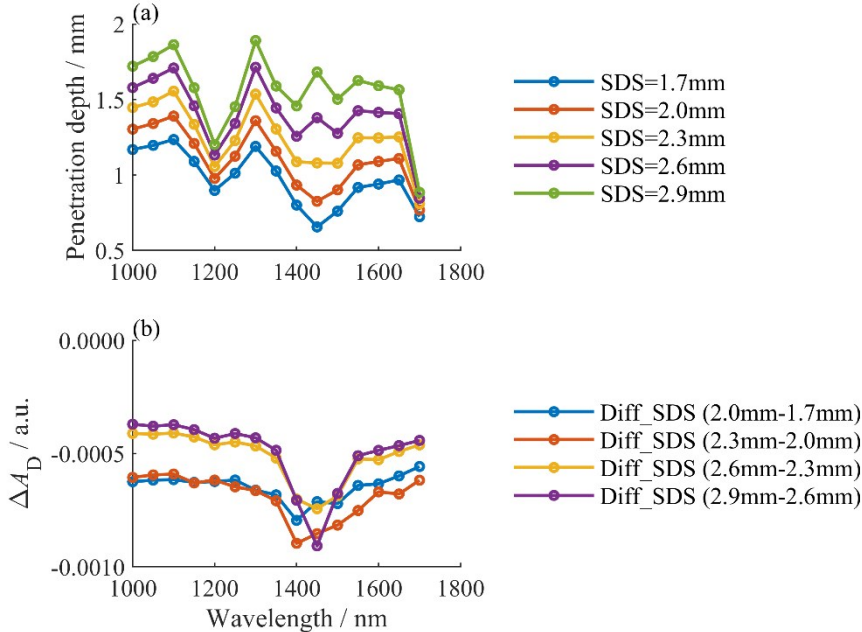


图2 三层皮肤组织模拟结果 (a) 平均穿透深度; (b) 1 mmol/L 葡萄糖引起的差分吸光度变化量

Fig.2 Simulation results of the three-layer skin tissue model. (a): Mean penetration depth; (b): Change in differential absorbance induced by 1 mmol/L glucose.

1.2 激光多普勒血流测量

LDF 是一种基于多普勒频移原理、用于无创检测组织微循环灌注的技术。在人体皮肤组织中,微血管内红细胞处于相对运动状态,而周围组织结构相对静止。探头发出的低功率激光进入皮肤后,在组织内发生多次吸收和散射。其中,被静止组织散射的光频率基本保持不变,而被运动红细胞散射的光会因红细胞相对于探测器的运动产生微小频率偏移,即多普勒频移。该频移量及其信号强度与红细胞运动速度和运动红细胞数量有关,因此可用于估计局部皮肤微循环灌注状态^[23]。

血流量(Flux, PU)是 LDF 中最常用的微循环灌注综合指标,主要反映采样体积内运动红细胞浓度与其平均运动速度的共同变化。其近似计算公式如下:

$$\text{Flux} \approx \text{Conc} \times \text{Speed} \quad (3)$$

其中 Conc 表征运动的红细胞浓度,Speed 表征红细胞平均运动速度。

本文采用 Moor Instruments VMS-LDF1(Moor Instruments, UK)及 VP1/7 型皮肤接触式探头(图 3)监测皮肤微循环灌注变化。光源波长为 785 nm,该探头的入射光纤与接收光纤间距为 0.5 mm,信号处理带宽为 15 kHz。根据探头结构及光纤间距,其平均探测深度约为 1 mm。本研究所获得的激光多普勒血流信号主要反映真皮浅层乳头下血管丛附近的微循环灌注变化。

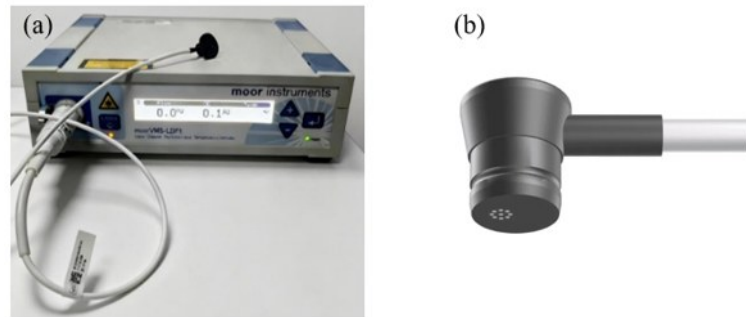


图3 本文采用的LDF (a) 血流仪;(b) 探头

Fig.3 Laser Doppler flowmeter (LDF) used in this study (a): Flowmeter; (b): Probe.

1.3 人体实验设置

近红外漫反射光谱法测量实验系统如图 4 所示,主要由光源、探测器、光开关以及数据采集处理单元构成。光源模块包括 6 个超辐射发光二极管,中心波长依次为 1050 nm、1219 nm、1314 nm、1409 nm、1550 nm 和 1609 nm,并具有对应的 3 dB 带宽。探测模块采用多环铟镓砷检测器,该检测器由 5 个独立同心环形探测单元组成,各环宽度均为 0.2 mm,并与入射光纤同轴排布,可分别获得 SDS 为 1.7、2.0、2.3、2.6 和 2.9 mm 的漫反射信号。本文选取 SDS 为 2.6 mm 和 2.9 mm 条件下的差分吸光度值作为光谱特征数据。该差分信号主要反映真皮层附近组织的光学响应,可在一定程度上削弱浅表组织干扰,因此用于后续光谱分析^[7,24]。

此外,为降低探头-皮肤界面变化引起的测量误差,本研究建立了标准化的界面控制策略^[7,19],包括:(1)利用亲肤型无纺布环形双面胶带固定探头,减少测量过程中探头位移;(2)通过双十字激光定位技术实现姿态的重复定位,保证光路耦合条件的一致性;(3)在探头与皮肤之间设置微小空气间隙,避免汗液积聚导致界面光学性质变化;(4)在采集正式数据前保持探头佩戴时间不少于 10 min,以促进接触压力和组织状态达到稳定。

共 6 名健康受试者参与光谱及生理指标测量实验,受试者基本特征见表 1。所有受试者均已签署知情同意书,本研究方案已通过天津大学伦理委员会审批。实验前,所有受试者均保持空腹状态。实验过程中,环境温度控制在 25-26℃,测量部位选取小臂伸侧。传感器温度设置为高于基础皮肤温度 0.5℃。光谱采集采用间歇测量模式,每隔约 5 min 采集一次,每次采集时间约为 30 s。测量过程中采用双十字激光定位,以确保不同测量时刻受试者姿势及测量位置的一致性和可重复性^[7]。实验过程中采用血糖仪(Accu-Chek, 瑞士)测量指尖血糖浓度。为使葡萄糖负荷实验和乳清蛋白负荷实验诱导机体产生相近水平的胰岛素反

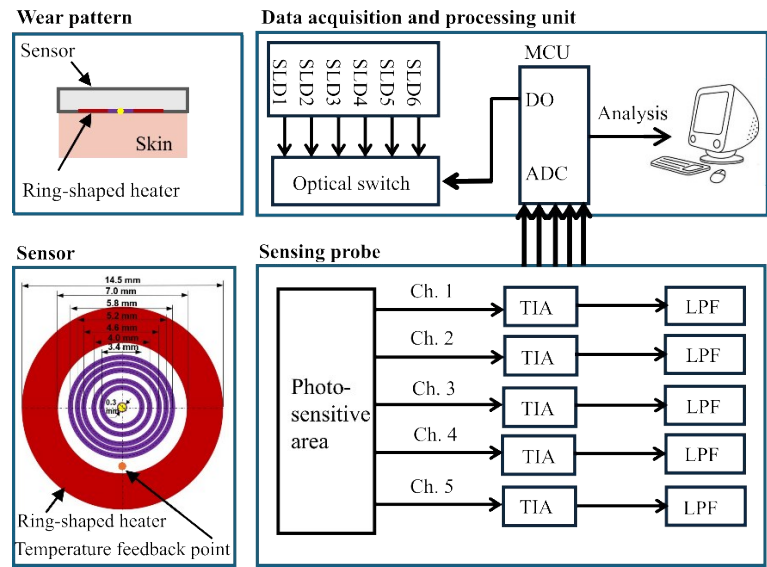


图4 实验系统设置
Fig. 4 Experimental system setup

应,本研究将两种负荷物质的摄入量均设定为 20 g。

实验共分为三大部分。(1)实验 1 为空腹评估实验,受试者 1 和受试者 2 佩戴传感器,连续监测 1 h 内的光谱变化。(2)实验 2 为葡萄糖负荷实验,6 名受试者在光谱信号稳定后摄入 100 mL 含 20 g 葡萄糖的水溶液,并连续监测摄入后的光谱变化。进一步,受试者 1 和受试者 2 另再开展一次葡萄糖负荷实验。受试者 1 和受试者 2 在同一测量部位另行开展 LDF 测量。(3)实验 3 为乳清蛋白负荷实验,6 名受试者在光谱信号稳定后摄入 100 mL 含 20 g 乳清蛋白(AIDOKE,新西兰)的水溶液,并连续监测摄入后的光谱变化。该乳清蛋白粉的营养成分组成如下:每 100 g 产品中含蛋白质 92.0 g,碳水化合物 0 g,钠 539 mg。其中,受试者 1 和受试者 2 在同一测量部位另行开展 LDF 测量。此外,受试者 1 和受试者 2 分别在两次负荷实验期间间隔约 30 min 进行静脉采血,用于生化检测血清胰岛素浓度。

表 1 受试者的人口统计学和基本特征
Table 1 Demographic and baseline characteristics of the subjects

Subject	Age	Sex (Male/Female)	BMI (kg/m ²)	Fitzpatrick Skin Type
#1	38	Male	24.9	III
#2	35	Male	23.0	III
#3	25	Male	28.1	III
#4	47	Female	24.3	III
#5	27	Male	25.5	III
#6	34	Male	18.6	III

2 结果

2.1 胰岛素浓度测量结果

在葡萄糖负荷实验和乳清蛋白负荷实验中,分别于摄入时刻和摄入后 30 min 采集受试者 1 与受试者 2 的静脉血样,并检测其血清胰岛素浓度,结果如表 2 所示。两名受试者摄入 20 g 葡萄糖后 30 min,胰岛素浓度平均升高 29.61 μ U/mL;摄入 20 g 乳清蛋白后 30 min,胰岛素浓度平均升高 19.82 μ U/mL。结果表明,两名受试者在葡萄糖负荷和乳清蛋白负荷后 30 min 的血清胰岛素浓度均较基线升高,其中葡萄糖负荷在 30 min 时的胰岛素升高幅度约为乳清蛋白负荷的 1.49 倍。乳清蛋白负荷虽略低于葡萄糖负荷,但仍产生了明显的胰岛素分泌效应。另外,胰岛素分泌水平受个体基础状态、胰岛素敏感性等多因素有关。

表2 摄入20 g葡萄糖及乳清蛋白前后生理指标测定结果

Table 2 Physiological measurements before and after ingestion of 20 g glucose and whey protein

Subject	Glucose load- ing—Insulin at 0 min ($\mu\text{U}/\text{mL}$)	Glucose load- ing—Insulin at 30 min ($\mu\text{U}/\text{mL}$)	Insulin change ($\mu\text{U}/\text{mL}$)	Whey protein loading—Insulin at 0 min ($\mu\text{U}/\text{mL}$)	Whey protein loading—Insulin at 30 min ($\mu\text{U}/\text{mL}$)	Insulin change ($\mu\text{U}/\text{mL}$)
#1	5.91	38.31	32.4	17	38	21
#2	12.05	38.86	26.81	4.66	23.3	18.64

2.2 激光多普勒血流仪测量结果

受试者1与受试者2分别进行了葡萄糖负荷和乳清蛋白负荷期间的LDF测量实验,结果如图5和图6所示。在葡萄糖负荷期间(摄入后30 min内),两名受试者的Flux变化量分别为3.9 PU和-5.6 PU;Conc变化量分别为7.6 AU和-6.9 AU。在乳清蛋白负荷期间(摄入后30 min内),两名受试者的Flux变化量分别为1.0 PU和-1.0 PU;Conc变化量分别为2.7 AU和-4.0 AU。需要说明的是,受血流脉动和皮肤自身抖动影响,血流仪数据本身存在一定的不确定度,如Flux在5分钟时间窗口下的波动量(标准差)约0.6~2.5 PU,而Conc的波动量约2.6~5.7 AU,而进食乳清蛋白或者葡萄糖后观察Flux和Conc的变化需大于3倍的仪器自身波动,才被认为发生了显著变化。因此,与仪器自身波动相比,两名受试者在葡萄糖负荷和乳清蛋白负荷期间皮肤微循环血流相关指标(Flux和Conc)的变化幅度均不显著,且变化方向有随机性,推测这些变化可能与环境因素及个体生理状态的随机波动有关。

2.3 近红外漫反射光谱测量结果

受试者1与受试者2的长期空腹评估实验结果如图7所示。虽然本研究采用差分吸光度以减少浅表

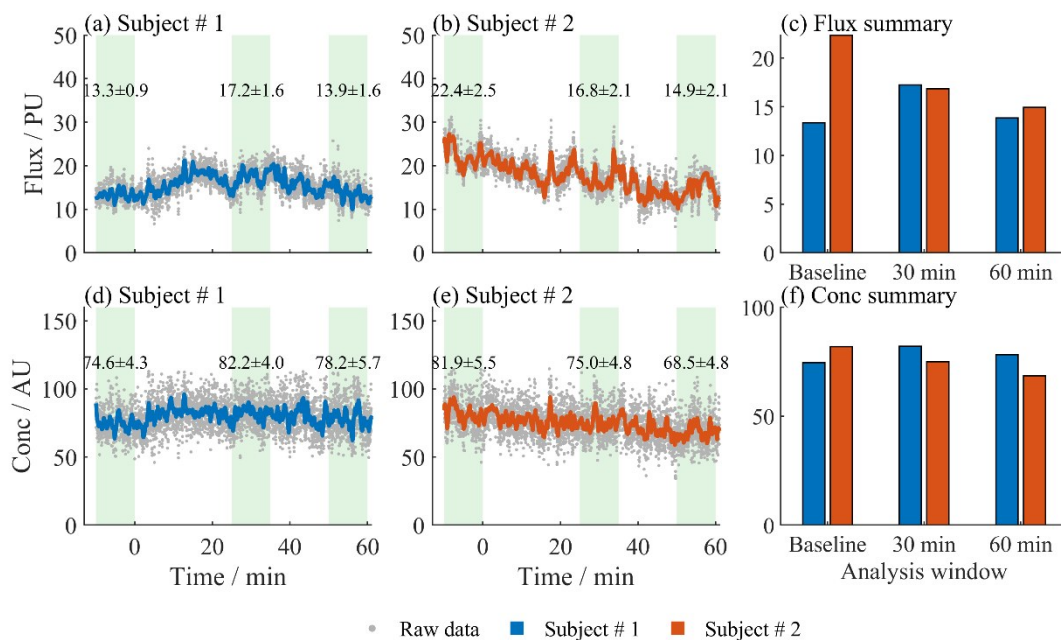


图5 葡萄糖负荷过程中LDF测量结果(0 min时摄入100 mL含20 g葡萄糖的水溶液)。(a)和(d)分别为受试者1的Flux和Conc变化结果;(b)和(e)分别为受试者2的Flux和Conc变化结果;(c)和(f)分别为两名受试者在选定5 min时间窗口内Flux和Conc测量值的平均结果。注:绿色阴影区域表示用于计算平均Flux和Conc的5 min时间窗口,区域内标注数值分别表示该时间窗口内测量数据的均值及标准差。

Fig. 5 LDF measurement results during glucose loading (100 mL aqueous solution containing 20 g glucose was orally ingested at 0 min). (a) and (d) show the Flux and Conc variations of subject 1, respectively; (b) and (e) show the Flux and Conc variations of subject 2, respectively; (c) and (f) present the mean values of Flux and Conc measurements for the two subjects within the selected 5 min time window. Note: The green shaded region indicates the 5 min time window used for calculating the average Flux and Conc values. The values labeled within the shaded region represent the mean and standard deviation of the measurements during the corresponding time interval.

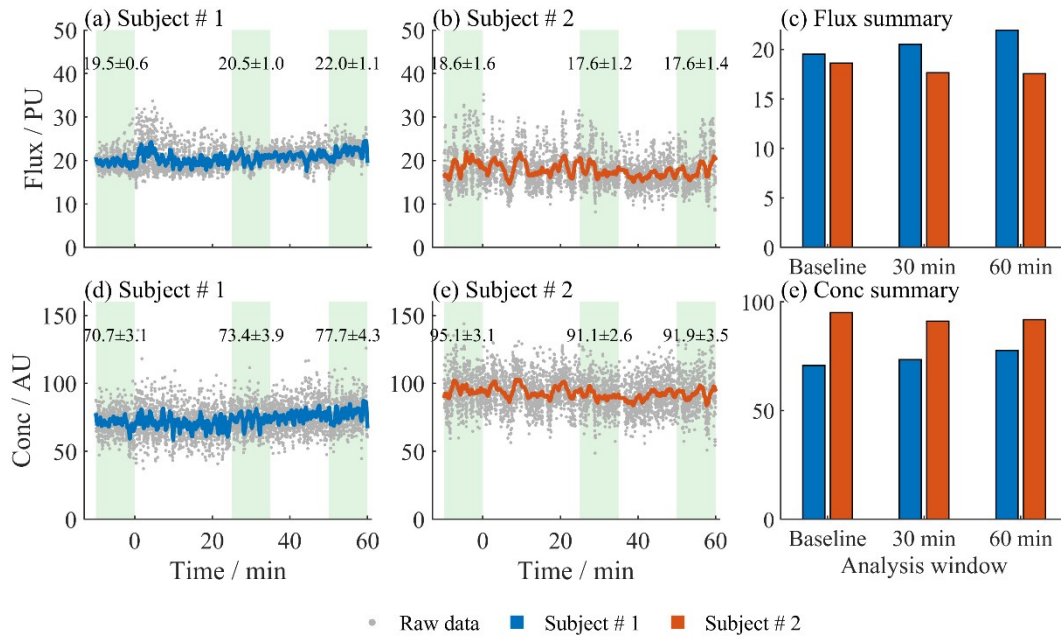


图6 乳清蛋白负荷过程中LDF测量结果(0 min时摄入100 mL含20 g乳清蛋白的水溶液)。(a)和(d)分别为受试者1的Flux和Conc变化结果;(b)和(e)分别为受试者2的Flux和Conc变化结果;(c)和(f)分别为两名受试者在选定5 min时间窗口内Flux和Conc测量值的平均结果。注:绿色阴影区域表示用于计算平均Flux和Conc的5 min时间窗口,区域内标注数值分别表示该时间窗口内测量数据的均值及标准差。

Fig. 6 LDF measurement results during whey protein loading (100 mL aqueous solution containing 20 g whey protein was orally ingested at 0 min). (a) and (d) show the Flux and Conc variations of subject 1, respectively; (b) and (e) show the Flux and Conc variations of subject 2, respectively; (c) and (f) present the mean values of Flux and Conc measurements for the two subjects within the selected 5 min time window. Note: The green shaded region indicates the 5 min time window used for calculating the average Flux and Conc values. The values labeled within the shaded region represent the mean and standard deviation of the measurements during the corresponding time interval.

组织和系统漂移的影响,但光谱信号仍可能受到受试者重置、传感器接触压力变化等多因素影响。尤其是在重置测量姿势时,皮肤与传感器接触界面的微小差异可能导致光路变化,从而引起光谱基线波动。结果显示,受试者1和受试者2的1550 nm差分吸光度标准差分别约为0.0005 a.u.和0.0003 a.u.。根据Ge等人^[8]报道的人体血糖检测灵敏度,在灵敏度较高的1550 nm波长处,采用SDS为2.6 mm与2.9 mm进行差分,1 mmol/L血糖浓度变化约对应0.001 a.u.的差分吸光度变化。由此可估算本研究所用传感器的极限检测精度约为0.4 mmol/L^[19]。该结果可作为后续葡萄糖负荷实验中光谱变化可检测性的参考依据。

6名受试者的葡萄糖负荷及乳清蛋白负荷实验结果如图8和图9所示。图8的葡萄糖负荷实验结果显示,6名受试者的8例实验中近红外光谱信号均出现与血糖浓度变化趋势较一致(负相关)的响应。其中,1550 nm波长处差分吸光度随血糖变化的灵敏度最高。图8中的|R|为3~4次采血时刻测得的1550 nm差分吸光度与血糖值间的相关系数绝对值。可以看出8例实验中|R|分布在0.66~0.98之间,平均|R|为0.82。可见,不同次实验间相关系数|R|存在一定差异。其中受试者1进行了两次实验(图8(a)和8(g)),两次实验|R|也不同,分别为0.92和0.69,受试者2也进行了两次实验(图8(b)和8(h)),两次实验|R|为0.66和0.78。上述结果说明除血糖浓度变化外,实验过程中可能伴随了人体生理状态的变化,这些变化使得信号缓慢向上或者向下漂移,而产生这些变化的原因可能是多样的,其与个体差异有关,也与个体在不同天的生理状态有关。相比之下,在乳清蛋白负荷期间,所有受试者的血糖浓度均未出现显著变化,光谱信号整体也较为稳定。因此,两种负荷条件下的光谱信号表现出明显差异。图10展示了受试者在两种负荷实验后30 min内差分吸光度变化量的对比结果。图中星号表示葡萄糖负荷与乳清蛋白负荷后30 min内差分吸光度变化量之间存在统计学差异。采用配对t检验比较受试者在两种负荷条件下各波长差分吸光度变化量的差异。结果显示,所有波长均达到统计学显著水平($p < 0.05$),表明葡萄糖负荷与乳清蛋白负荷条件下的差分吸光度变化量存在显著差异。

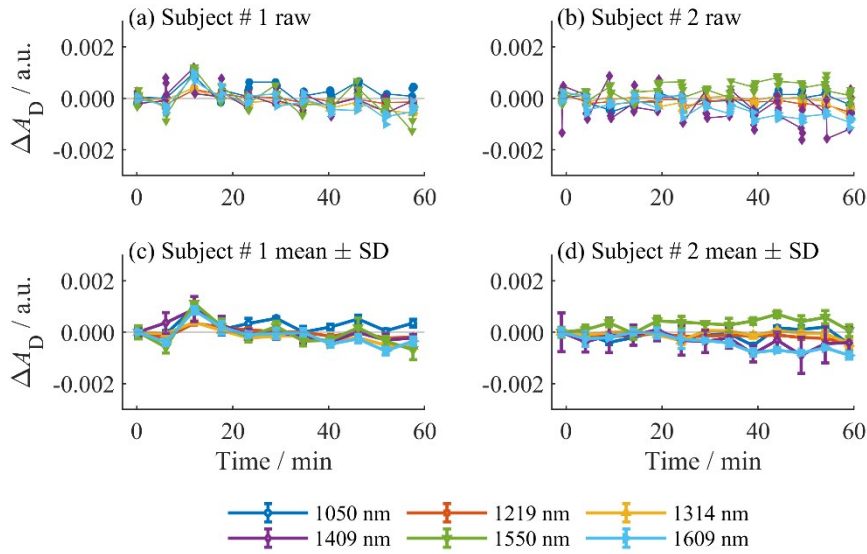


图7 空腹评估实验 (a)和(c)为受试者1;(b)和(d)为受试者2

Fig. 7 Fasting assessment experiment. (a) and (c) show Subject 1, while (b) and (d) show Subject 2.

3 讨论

本研究通过对比葡萄糖负荷实验和乳清蛋白负荷实验,结合 LDF 和近红外漫反射光谱技术,分别监测皮肤真皮浅层微循环血流变化及局部皮肤组织光学响应,初步探讨了胰岛素浓度升高对真皮浅层微循环血流及皮肤近红外光谱信号(1050 - 1609 nm 波段)的响应。在本研究条件下的实验结果表明,乳清蛋白负荷诱导的胰岛素分泌增加对真皮浅层微循环血流的影响较小,其对所采集的 1050 - 1609 nm 波段的光谱信号影响也较小,因此其可能不是降低血糖测量精度的主要干扰源。此外,由 6 名受试者完成的 8 例葡萄糖负荷实验结果表明,1550 nm 单波长光信号与血糖变化均表现出较高的相关性,相关系数分布于 0.66 ~ 0.98 之间。从 2 名受试者的重复实验结果还可以看出,同一受试者不同次实验的相关系数也存在波动,时高时低。导致相关系数波动的具体原因仍有待进一步研究。

首先,LDF 信号受光纤间距和光源-探测器距离影响,不同距离对应不同探测深度。本研究所采用的 LDF 测量深度在 1 mm 附近,主要反映真皮浅层乳头下血管丛附近的微循环水平。已有研究表明,胰岛素诱导的毛细血管募集更多发生于骨骼肌等深层代谢活跃的组织中,对真皮浅层微循环的影响可能较小^[25-27]。此外,受试者局部温度、探头压力、测量部位、皮肤接触状态和仪器自身精度水平等因素也可能影响 LDF 结果。本文对两位受试者做了 LDF 测量,变化量均不显著。结合已有文献及本实验观察结果,本文认为两位受试者胰岛素分泌后对皮肤浅层血流的影响可能相对有限。

其次,部分受试者在乳清蛋白负荷后的近红外光谱信号波动性有所增加,其波动水平略高于空腹状态下的信号波动水平。尤其是受试者 5 在 1050 nm 波长处的差分吸光度信号波动较为明显,而该波长对血红蛋白吸收较为敏感,提示该受试者在摄入乳清蛋白后可能存在一定程度的微循环波动。因此,胰岛素升高可能诱发轻微的皮肤血流灌注变化。然而,对比 6 名受试者葡萄糖负荷与乳清蛋白负荷后的光谱响应可以发现,摄入葡萄糖水后引起的信号变化幅度明显大于摄入乳清蛋白后的信号变化幅度。这说明,胰岛素诱发的微循环血流变化即使存在,其影响也相对有限,特别是对血糖变化最灵敏的 1550 nm 处也未观察到稳定且可分辨的变化。

本研究仍属于初步探索性研究。受试者数量较少,光谱实验仅纳入 6 名健康受试者,血清胰岛素浓度检测及 LDF 测量实验仅纳入 2 名健康受试者,且暂未开展糖尿病患者相关测试,因此实验结果仍具有一定局限性。另外,葡萄糖负荷后,近红外光谱信号不仅受到血糖浓度变化的影响,还可能受到胰岛素分泌、微循环血流灌注、组织水分分布以及代谢状态变化等多种生理因素的共同影响^[28-30]。由于血糖升高和下降阶段对应的机体调节过程可能存在差异,且部分生理响应可能存在时间延迟,因此在血糖恢复阶段,光谱信号

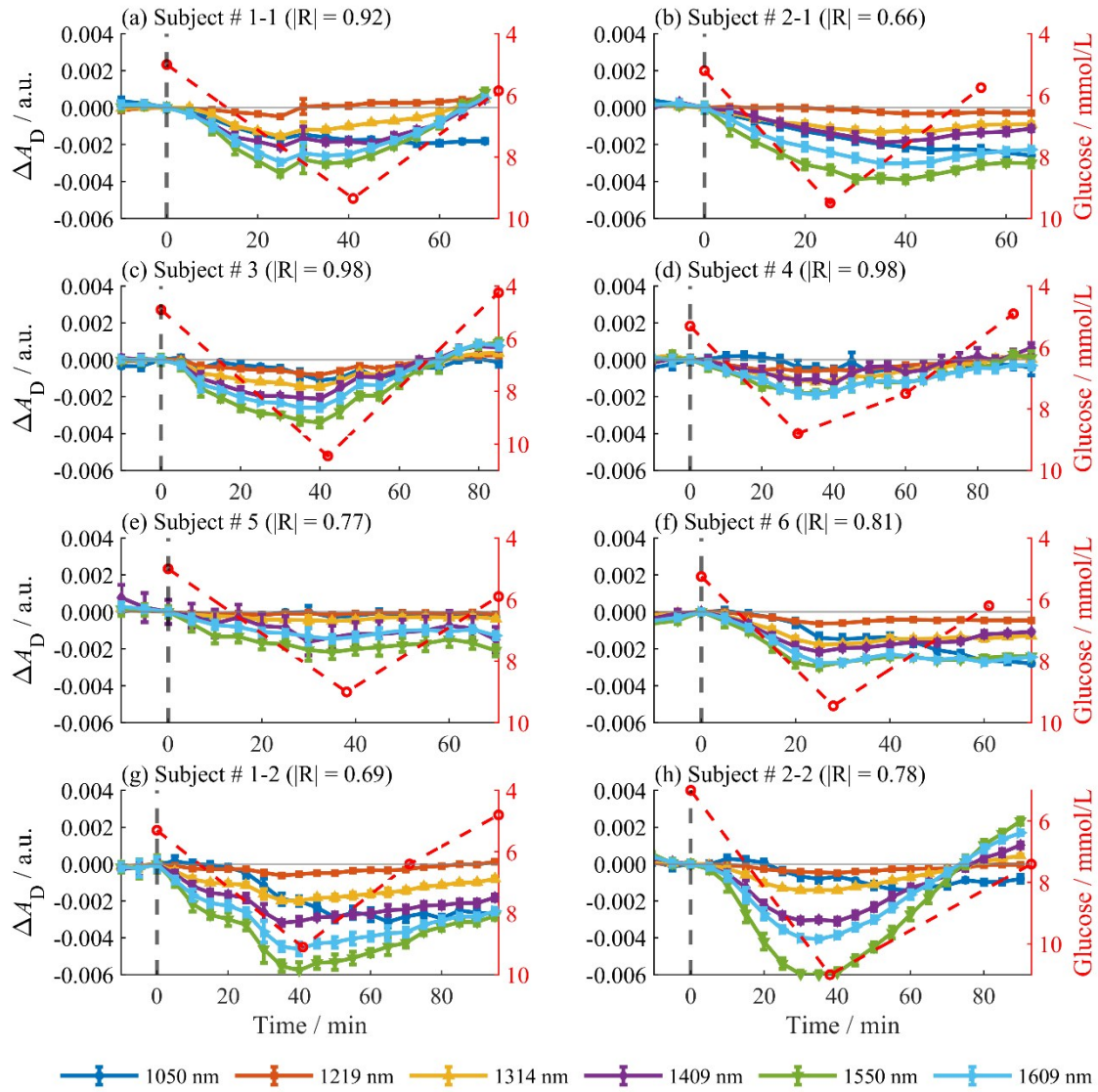


图8 葡萄糖负荷期间光谱测量结果及1550 nm光信号与血糖的相关系数(0 min时摄入100 mL含20 g葡萄糖的水溶液)(a) - (f)分别对应受试者1-6的光谱变化结果;(g)和(h)分别为受试者1和2的重复实验结果。注:图中 $|R|$ 为1550 nm差分光度与血糖值间的相关系数绝对值。

Fig. 8 Spectral measurement results during glucose loading and correlation coefficients between the 1550 nm optical signal and blood glucose concentration (100 mL aqueous solution containing 20 g glucose was orally ingested at 0 min). (a) - (f) Spectral variation results corresponding to subjects 1 - 6, respectively; (g) and (h) Repeatability test results of subjects 1 and 2, respectively. Note: $|R|$ represents the absolute value of the correlation coefficient between the differential absorbance at 1550 nm and blood glucose concentration.

不一定能够完全同步反映血糖变化。此外,不同受试者之间以及同一受试者不同实验过程中的生理响应差异,也可能导致光谱信号与血糖变化之间相关性的变化。本文对血糖相关光谱信号来源的解释尚不充分,不能完全排除其他生理因素对光谱信号的影响。后续研究将进一步扩大样本量,并同步监测更多生理指标,以明确干扰的具体来源。

4 结论

本研究通过对比葡萄糖负荷实验和乳清蛋白负荷实验,结合LDF和近红外漫反射光谱,初步探讨了胰岛素浓度升高对真皮浅层微循环血流及皮肤近红外光谱信号(1050 - 1609 nm波段)的影响。胰岛素检测结果表明,两名受试者在葡萄糖负荷和乳清蛋白负荷后30 min的血清胰岛素浓度均较基线升高,约为基线的2.24倍及5倍。实验结果显示,葡萄糖负荷后,6名受试者的近红外光谱信号均出现与血糖浓度变化趋势较

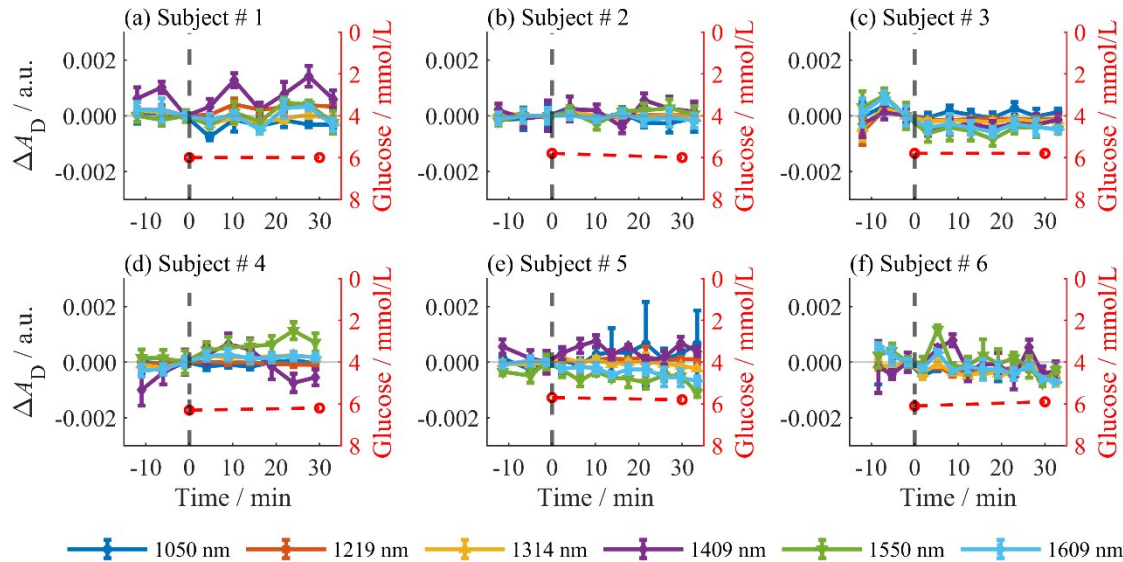


图9 乳清蛋白负荷期间光谱测量结果(0 min时摄入100 mL含20 g乳清蛋白的水溶液)(a) - (f) 受试者1-6
Fig. 9 Spectral measurement results during whey protein loading. (Subjects ingested 100 mL of water containing 20 g whey protein at 0 min) (a) - (f): Subjects 1 - 6.

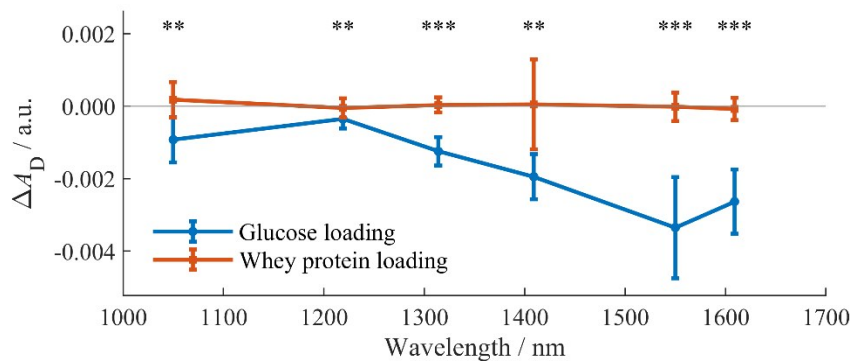


图10 葡萄糖负荷与乳清蛋白负荷后30 min内差分吸光度变化量的对比(样本量 $n = 6$)。图中*, **和***分别表示两组数据间差异具有统计学意义($p < 0.05$)、高度统计学意义($p < 0.01$)和极高度统计学意义($p < 0.001$)。
Fig. 10 Comparison of changes in differential absorbance within 30 min after glucose and whey protein loading ($n = 6$). The symbols *, **, and *** indicate statistically significant differences between the two groups at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively.

一致的响应;而乳清蛋白负荷后光谱信号整体较为平稳。两种负荷实验后30 min内差分吸光度变化量存在显著差异($p < 0.05$)。2名受试者的LDF测量结果显示,葡萄糖负荷和乳清蛋白负荷期间皮肤微循环相关指标变化幅度均较小,且变化方向有随机性。综上,在本研究条件下,乳清蛋白负荷下的胰岛素浓度升高对真皮浅层微循环及1050-1609 nm波段光谱信号的影响相对有限。

参考文献

- [1] KOHL M, ESSENPREIS M, COPE M. The influence of glucose concentration upon the transport of light in tissue-simulating phantoms[J]. *Physics in Medicine and Biology*, 1995, 40(7): 1267-1287.
- [2] RAMBLA F J, GARRIGUES S, De La Guardia M. PLS-NIR determination of total sugar, glucose, fructose and sucrose in aqueous solutions of fruit juices[J]. *Analytica Chimica Acta*, 1997, 344(1-2): 41-53.
- [3] RORSMAN P, ASHCROFT F M. Pancreatic β -Cell Electrical Activity and Insulin Secretion: Of Mice and Men[J]. *Physiological Reviews*, 2018, 98(1): 117-214.
- [4] SALTIEL A R, KAHN C R. Insulin signalling and the regulation of glucose and lipid metabolism[J]. *Nature*, 2001, 414(6865): 799-806.
- [5] LETO D, SALTIEL A R. Regulation of glucose transport by insulin: traffic control of GLUT4[J]. *Nature Reviews Molecular Cell Biology*, 2012, 13(6): 383-396.

- [6] SERNÉ E H, IJZERMAN R G, GANS R O B, et al. Direct Evidence for Insulin-Induced Capillary Recruitment in Skin of Healthy Subjects During Physiological Hyperinsulinemia[J]. *Diabetes*, 2002, 51(5): 1515-1522.
- [7] HAN T, LIU J, LIU R, et al. In Vivo Near-Infrared Noninvasive Glucose Measurement and Detection in Humans[J]. *Applied Spectroscopy*, 2022, 76(9): 1100-1111.
- [8] GE Q, HAN T, LIU R, et al. Evaluation and Validation on Sensitivity of Near-Infrared Diffuse Reflectance in Non-Invasive Human Blood Glucose Measurement[J]. *Sensors*, 2024, 24(18): 5879.
- [9] SALEHI A, GUNNERUD U, MUHAMMED S J, et al. The insulinogenic effect of whey protein is partially mediated by a direct effect of amino acids and GIP on β -cells[J]. *Nutrition & Metabolism*, 2012, 9(1): 48.
- [10] ANG T, MASON S A, DAO G M, et al. The impact of a single dose of whey protein on glucose flux and metabolite profiles in normoglycemic males: insights into glucagon and insulin biology[J]. *American Journal of Physiology-Endocrinology and Metabolism*, 2023, 325(6): E688-E699.
- [11] ŠORLI J, LENASI H. The Effect of Acute Hyperglycaemia Induced by Oral Glucose Load on Heart Rate Variability and Skin Microvascular Reactivity in Young Adults[J]. *Life*, 2023, 14(1): 56.
- [12] MEIJER R I, DE BOER M P, GROEN M R, et al. Insulin-Induced Microvascular Recruitment in Skin and Muscle are Related and Both are Associated with Whole-Body Glucose Uptake[J]. *Microcirculation*, 2012, 19(6): 494-500.
- [13] HOHBERG C, FORST T, LARBIG M, et al. Effect of Insulin Glulisine on Microvascular Blood Flow and Endothelial Function in the Postprandial State[J]. *Diabetes Care*, 2008, 31(5): 1021-1025.
- [14] CADUFF A, TALARY M S, ZAKHAROV P. Cutaneous Blood Perfusion as a Perturbing Factor for Noninvasive Glucose Monitoring[J]. *Diabetes Technology & Therapeutics*, 2010, 12(1): 1-9.
- [15] LEVICK J R, MICHEL C C. Microvascular fluid exchange and the revised Starling principle[J]. *Cardiovascular Research*, 2010, 87(2): 198-210.
- [16] BRIERS J D. Laser Doppler, speckle and related techniques for blood perfusion mapping and imaging[J]. *Physiological Measurement*, 2001, 22(4): R35-R66.
- [17] LIU W, CHEN J, LIU X, et al. Analysis on direct and additional temperature spectra of skin: A near-infrared diffuse reflectance spectroscopy study with temperature control[J]. *Infrared Physics & Technology*, 2026, 152: 106187.
- [18] LIU Wenbo, LIU Jin, HAN Tongshuai, et al. Simulation of the effect of dermal thickness on non-invasive blood glucose measurement by near-infrared spectroscopy [J]. *Spectroscopy and Spectral Analysis*, 2023, 43(9): 2699-2704.
刘文博, 刘瑾, 韩同帅, 等. 皮肤真皮层厚度对近红外光谱法无创血糖测量影响的仿真[J]. *光谱学与光谱分析*, 2023, 43(9): 2699-2704.
- [19] LIU W, HAN T, CHEN W, et al. Design Key Points of High-Performance Diffuse Reflectance Optical Sensors for Non-Invasive Blood Glucose Measurement[J]. *Sensors*, 2025, 25(4): 998.
- [20] ZHEREBTSOV E, DREMIN V, POPOV A, et al. Hyperspectral imaging of human skin aided by artificial neural networks[J]. *Biomedical Optics Express*, 2019, 10(7): 3545.
- [21] WILSON R H, NADEAU K P, JAWORSKI F B, et al. Review of short-wave infrared spectroscopy and imaging methods for biological tissue characterization[J]. *Journal of Biomedical Optics*, 2015, 20(3): 030901.
- [22] AMEROV A K, CHEN J, ARNOLD M A. Molar Absorptivities of Glucose and other Biological Molecules in Aqueous Solutions over the First Overtone and Combination Regions of the Near-Infrared Spectrum[J]. *Applied Spectroscopy*, 2004, 58(10): 1195-1204.
- [23] RAJAN V, VARGHESE B, VAN LEEUWEN T G, et al. Review of methodological developments in laser Doppler flowmetry[J]. *Lasers in Medical Science*, 2009, 24(2): 269-283.
- [24] GE Q, HAN T, LIU X, et al. Effects of Skin Blood Flow Fluctuations on Non-Invasive Glucose Measurement and a Feasible Blood Flow Control Method[J]. *Sensors*, 2025, 25(4): 1162.
- [25] JANSSON P, LARSSON A, LÖNNROTH P. Relationship between blood pressure, metabolic variables and blood flow in obese subjects with or without non-insulin-dependent diabetes mellitus[J]. *European Journal of Clinical Investigation*, 1998, 28(10): 813-818.
- [26] KARPE F, FIELDING B A, ILIC V, et al. Impaired Postprandial Adipose Tissue Blood Flow Response Is Related to Aspects of Insulin Sensitivity[J]. *Diabetes*, 2002, 51(8): 2467-2473.
- [27] KARPE F, FIELDING B A, ARDILOUZE J, et al. Effects of insulin on adipose tissue blood flow in man[J]. *The Journal of Physiology*, 2002, 540(3): 1087-1093.
- [28] SHEN Y, BU J, YU L, et al. Optical coherence tomography angiography allows longitudinal monitoring of angiogenesis in the critical-sized defect model[J]. *Frontiers in Physics*, 2023, 11: 1218179.
- [29] BASHKATOV A N, KOROLEVICH A N, TUCHIN V V, et al. In vivo investigation of human skin optical clearing and blood microcirculation under the action of glucose solution[J]. *Asian Journal of Physics*, 2006, 15(1): 1-14.
- [30] SILVA H, ŠORLI J, LENASI H. Oral Glucose Load and Human Cutaneous Microcirculation: An Insight into Flowmotion Assessed by Wavelet Transform[J]. *Biology*, 2021, 10(10): 953.

Comparison of Near-Infrared Diffuse Reflectance Spectral Responses and Skin Microcirculatory Changes Under Glucose and Whey Protein Loading

LIU Wenbo¹, YAO Mingfei², CHEN Jiayu¹, CHEN Yuheng¹, SUN Di², LIU Jin¹, HAN Tongshuai¹

(1 *State Key Laboratory of Precision Measurement Technology and Instruments, Tianjin University, Tianjin 300072, China*)

(2 *Sunrise Technology Co., Ltd., Tianjin 300192, China*)

Abstract: This study aimed to evaluate whether insulin elevation affects superficial dermal microcirculatory blood flow and near-infrared diffuse reflectance spectral signals within the 1050 – 1609 nm wavelength range. To separate insulin-related effects from blood glucose-related effects, a whey protein load experiment was designed and compared with a glucose load experiment. By combining serum insulin measurement, laser Doppler flowmetry, and near-infrared diffuse reflectance spectroscopy, this study investigated whether insulin-induced superficial dermal perfusion changes could be detected optically and whether they should be considered a major physiological interference source in noninvasive blood glucose measurement. Six healthy volunteers were enrolled in the load experiments. Each subject ingested 100 mL of aqueous solution containing either 20 g glucose or 20 g whey protein. The glucose load was used to induce blood glucose elevation accompanied by insulin secretion, whereas the whey protein load was used to stimulate insulin secretion while minimizing blood glucose fluctuation. Near-infrared diffuse reflectance spectra were collected from the skin during both load conditions to monitor local optical responses in the superficial dermal region. Signals were acquired at six selected wavelengths: 1050, 1219, 1314, 1409, 1550, and 1609 nm, covering optical information related to hemoglobin, lipid, water, glucose, and tissue scattering. Among them, 1050 nm was considered relatively sensitive to hemoglobin absorption, while 1550 nm was regarded as the most sensitive wavelength for glucose-related spectral changes in this study. Differential absorbance was used as the main spectral feature. The spectral responses after glucose and whey protein loading were compared to determine whether insulin-related perfusion changes could produce distinguishable optical variations. In two of the six subjects, venous blood samples were collected at ingestion and 30 min after ingestion in both load experiments, and serum insulin concentration was measured by biochemical analysis. In the same two subjects, superficial dermal microcirculatory blood flow was monitored using laser Doppler flowmetry. The measurement depth of LDF was approximately 1 mm, mainly corresponding to the superficial dermis and the region near the subpapillary vascular plexus. The LDF signal was used to evaluate local microcirculatory perfusion because it reflects the combined contribution of moving red blood cell concentration and mean velocity within the sampling volume. In addition, the differential absorbance changes within 30 min after ingestion were statistically compared between the two load conditions. Serum insulin measurements showed that both glucose loading and whey protein loading clearly stimulated insulin secretion in the monitored subjects, indicating that the whey protein intervention successfully produced an insulin-elevating condition. However, LDF results showed that the superficial dermal microcirculatory blood flow signals did not change obviously during either load condition. The changes in LDF-related parameters were small during both experiments, and their magnitudes were comparable. This suggests that insulin elevation induced by the two interventions had a limited influence on superficial dermal microcirculatory perfusion under the present experimental conditions. The near-infrared spectral responses showed clear differences between the two load experiments. After glucose ingestion, the spectral signals of all six subjects changed noticeably and showed a trend generally consistent with blood glucose variation. Among the six selected wavelengths, the differential absorbance at 1550 nm showed relatively good correlation with blood glucose changes, suggesting that this wavelength was more sensitive to glucose-related optical responses. By contrast, after whey protein ingestion, the overall spectral signals remained relatively stable, and no significant systematic spectral change was observed. The differential absorbance changes within 30 min after ingestion were significantly different between the glucose load and whey protein load conditions, with $p < 0.05$, indicating that the glucose-induced spectral response was significantly stronger than the whey protein-induced response. Although the

overall spectral response after whey protein loading was weak, some subjects showed slightly increased spectral fluctuation compared with the fasting state. In particular, Subject 5 showed relatively obvious fluctuation in the 1050 nm differential absorbance signal. Since 1050 nm is relatively sensitive to hemoglobin absorption, this fluctuation may indicate a mild blood-related optical response or slight superficial microcirculatory variation after whey protein ingestion. However, compared with the glucose load experiment, the amplitude of spectral changes after whey protein ingestion was much smaller. Moreover, no stable or clearly distinguishable spectral variation was observed at 1550 nm after whey protein loading, although this wavelength showed the highest sensitivity to blood glucose variation during glucose loading. These results suggest that even if insulin-induced superficial dermal microcirculatory fluctuation exists, its influence on the collected 1050 – 1609 nm near-infrared spectral signals is limited and insufficient to generate a stable glucose-like spectral response. The results also indicate that the optical responses observed during glucose loading cannot be attributed mainly to insulin-related superficial dermal microcirculatory effects. Glucose loading may induce multiple physiological responses in addition to changes in blood glucose and insulin, including changes in cell size, local osmotic status, tissue metabolism, and other optical properties of skin tissue. Since this exploratory study included only six healthy subjects, and insulin concentration and LDF measurements were performed in only two subjects, the findings should be interpreted cautiously. Insulin elevation showed only a limited effect on superficial dermal microcirculatory blood flow and near-infrared diffuse reflectance spectral signals within the 1050 – 1609 nm range. Its influence was not sufficient to form a stable and distinguishable optical interference under the present experimental conditions.

Key words: Noninvasive blood glucose measurement; laser Doppler blood flow measurement; near-infrared diffuse reflectance spectroscopy; insulin; whey protein

OCIS Codes: 300.6340; 280.1415; 170.6935; 170.1470

CSTR: 32255.14.gzxb20275601.0130001