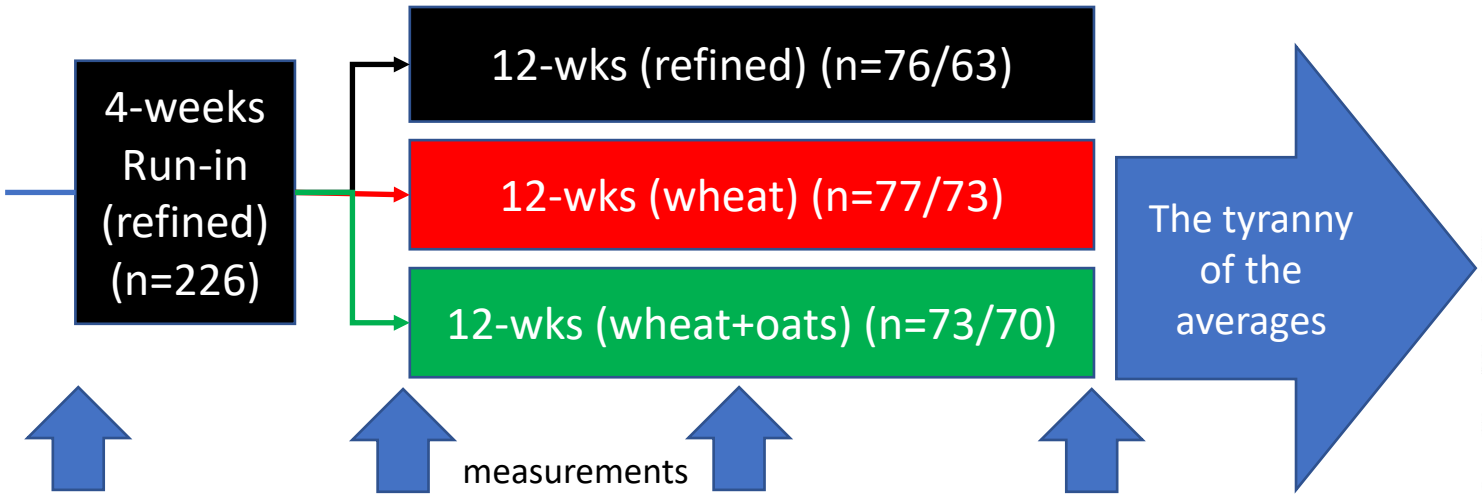
N-of-1 Methods in Precision Nutrition Research

Jose M Ordovas

JM-USDA-HNRCA at Tufts University

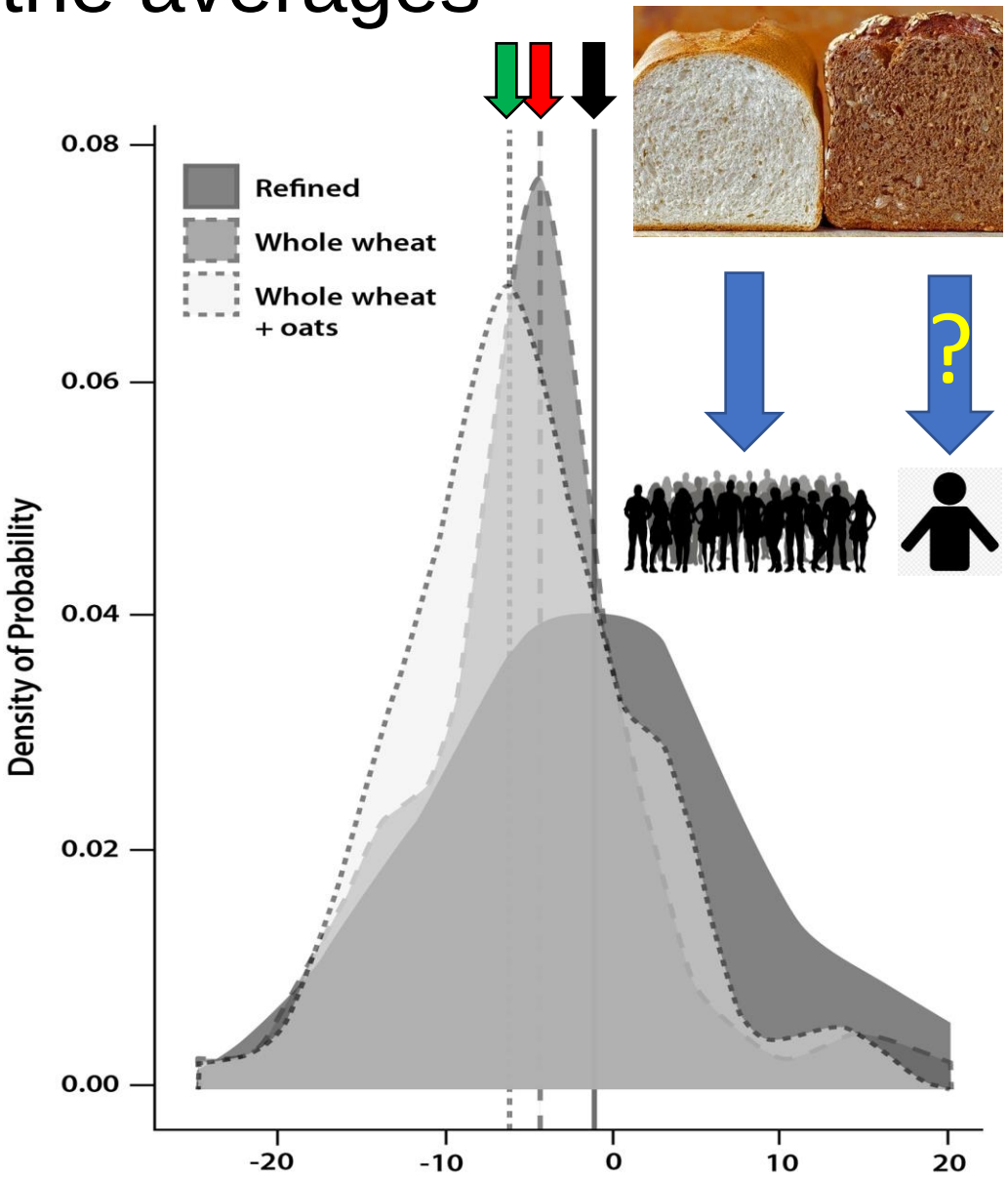
Nutrition trials: RCT and the tyranny of the averages

Effect of increased consumption of whole-grain foods on blood pressure and other cardiovascular risk markers in healthy middle-aged persons: an RCT



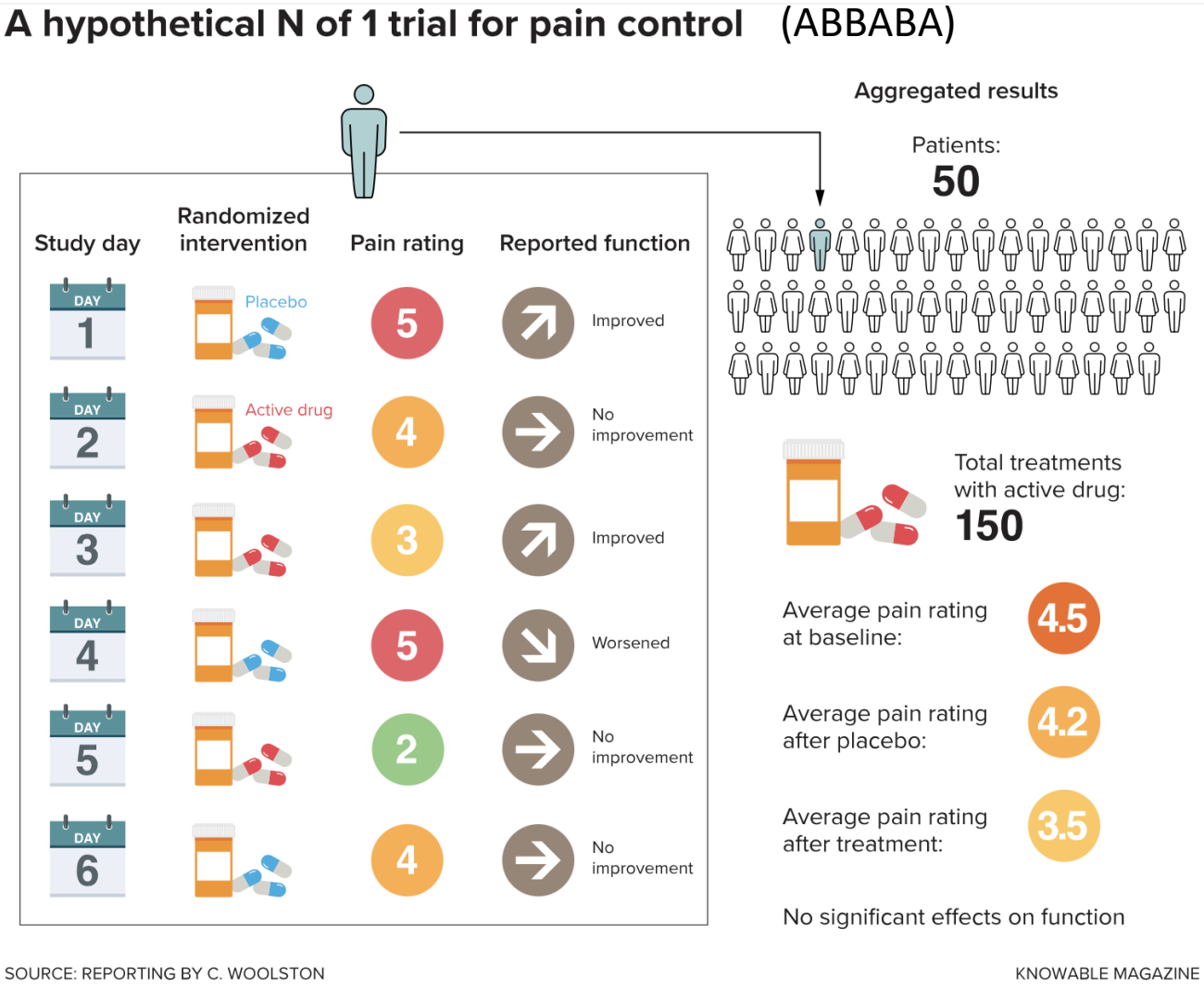
Conclusion: Daily consumption of 3 portions of whole-grain foods can **significantly reduce** cardiovascular disease risk in middle-aged people mainly through blood pressure–lowering mechanisms. The observed decrease in systolic blood pressure **could decrease the incidence of coronary artery disease and stroke** by ≥15% and 25%, respectively.

Tighe P, et al. Am J Clin Nutr. 2010;92(4):733-40.
Merlo J et al. SSM Popul Health. 2017;3:684-698.
Potter T, et al. Adv Nutr. 2021 Jun 1;12(3):579-589.



Distribution of difference in SBP between the start and end of the intervention across 3 dietary intervention groups

N-of-1 Trials: The Singular Science



For this patient, the drug outperformed the placebo for pain relief but the effect on function was less conclusive. At right, combining results from 50 individuals who followed the same procedure allows scientists to gauge the bigger picture: The drug shows promise for relieving pain relief but not for improving function.

Timeline n-of-1 trials (some highlights)

Richard Wiseman (1676), leg oedema and the laced stockings. An ABA N-of-1 design

Caleb Parry (1786). Formal, planned use of between two and six crossover periods of variable duration in 13 patients, to compare the purgative effects of three varieties of rhubarb.

John Haygarth (1800) did placebo-controlled trials to test the therapeutic claims made by Elisha Perkins for his ‘metallic tractors’.

Paul Martini (1932). Book setting out the methodological principles for assessing the effects of treatments.

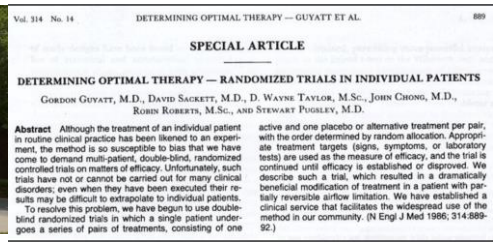
Brit. J. prev. soc. Med. (1953), 7, 163–179

THE SELF-CONTROLLED AND SELF-RECORDED CLINICAL TRIAL FOR LOW-GRADE MORBIDITY

BY

LANCELOT. HOGBEN and M. SIM

Birmingham



Vol. 314 No. 14 DETERMINING OPTIMAL THERAPY — GUYATT ET AL. 889

SPECIAL ARTICLE

DETERMINING OPTIMAL THERAPY — RANDOMIZED TRIALS IN INDIVIDUAL PATIENTS

GORDON GUYATT, M.D., DAVID SACKETT, M.D., D. WAYNE TAYLOR, M.SC., JOHN CHONG, M.D., ROBIN ROBERTS, M.SC., AND STEWART PUGSLEY, M.D.

Abstract Although the treatment of an individual patient in routine clinical practice has been likened to an experiment, the method is so susceptible to bias that we have come to demand multi-patient, double-blind, randomized controlled trials on matters of efficacy. Unfortunately, such trials have not or cannot be carried out for many clinical disorders; even when they have been executed their results may be difficult to extrapolate to individual patients.

To resolve this problem, we have begun to use double-blind randomized trials in which a single patient undergoes a series of pairs of treatments, consisting of one active and one placebo or alternative treatment per pair, with the order determined by random allocation. Appropriate treatment targets (signs, symptoms, or laboratory tests) are used as the measure of efficacy, and the trial is continued until efficacy is established or disproved. We describe such a trial, which resulted in a dramatically beneficial modification of treatment in a patient with partially reversible airflow limitation. We have established a clinical service that facilitates the widespread use of the method in our community. (N Engl J Med 1986; 314:889-92.)

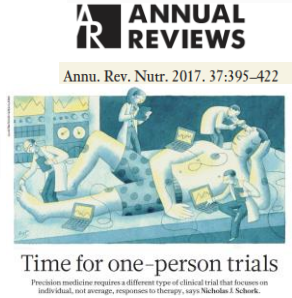
PERSPECTIVE

Adv Nutr 2021;12:579–589

Perspective: Application of N-of-1 Methods in Personalized Nutrition Research

Tilly Potter,[1] Rute Vieira,[2] and Baukje de Roos[1]

[1] *Rowett Institute, University of Aberdeen, United Kingdom; and* [2] *Institute of Applied Health Sciences, University of Aberdeen, United Kingdom*



ANNUAL REVIEWS

Annual Review of Nutrition

Annu. Rev. Nutr. 2017. 37:395–422

Single-Subject Studies in Translational Nutrition Research

Nicholas J. Schork[1,2,3] and Laura H. Goetz[2,4,5]

[1] Translational Genomics Research Institute, Phoenix, Arizona 85004; email: nschork@tgen.org

[2] J. Craig Venter Institute, La Jolla, California 92037; email: nschork@jcvi.org

[3] Departments of Psychiatry and Family Medicine and Public Health, University of California, San Diego, La Jolla, California 92037

[4] Department of Surgery, Scripps Clinic Medical Group, La Jolla, California 92037

[5] Department of Molecular and Experimental Medicine, The Scripps Research Institute, La Jolla, California 92037

Time for one-person trials

Precision medicine requires a different type of clinical trial that focuses on individual, not average, responses to therapy, says Nicholas J. Schork.

Schork NJ. Nature. 2015;520:609-11

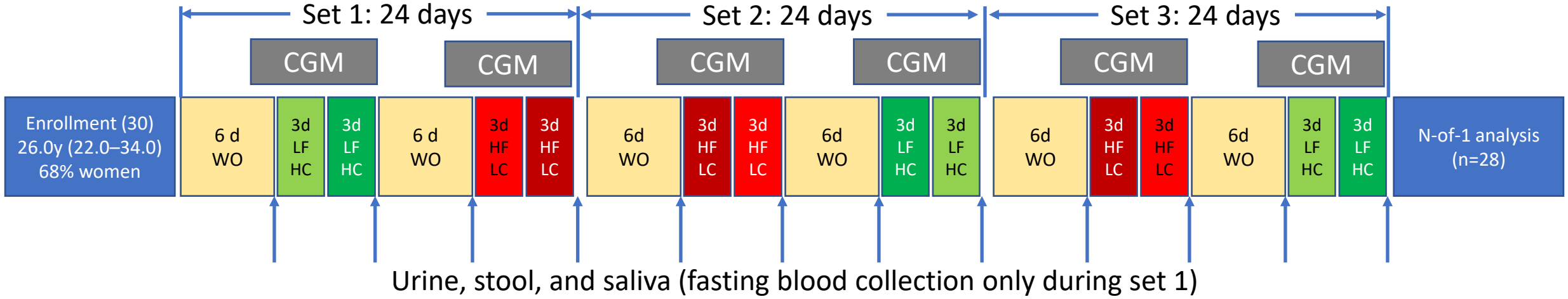
Ongoing and recently published N-of-1 trials relating to nutrition

Title of study/NIH listing	Study status	Participant description	Study design	Statistics	Intervention(s)/treatment(s)	Primary outcome(s)
Application of N-of-1 clinical trials in personalized nutrition research: a trial protocol for Westlake N-of-1 trials for macronutrient intake (NCT04125602)	Ongoing	Healthy participants	Repeated randomized (within-block) crossover n-of-1 (3 blocks	Bayesian models	High-fat, low-carbohydrate diet; Low-fat, high-carbohydrate diet	Postprandial glycemic responses, including postprandial maximum glucose and 0–24 h area under the curve
Coffee and real-time atrial and ventricular ectopy (CRAVE;NCT03671759)	Ongoing	Healthy participants	Interventional, randomized in 2-day blocks	Not stated	Caffeine consumption (versus withdrawal)	Change in cardiac ectopy burden (heart rhythm)
Measuring individual responses to a whole-grain and nuts intervention to reduce blood pressure in prehypertension (MI-DIET; NCT04326686)	Ongoing	Volunteers with mildly elevated blood pressure	Interventional ABA design	Dynamic modelling	DASH diet with whole grains and nuts provided	Adherence to intervention; Change in blood pressure levels
Personalized research on diet in ulcerative colitis and Crohn's disease (PRODUCE; NCT03301311)	Ongoing	Patients aged 7–18	Repeated, randomized crossover N-of-1	Individual and population-level analysis	Specific carbohydrate diet, Modified specific carbohydrate diet	Stool frequency and consistency; Pain interference; Gastrointestinal symptoms; Fecal Calprotectin
Personalized lifestyle intervention for improving functional health outcomes using N-of-1 tent-umbrella-bucket design (LIFE-HOUSE; NCT04005456)	Ongoing	Patients with a variety of chronic diseases	Crossover N-of-1 (depending on the clinical group)	Not stated	Varied interventions including dietary supplements, behavioral change support program, and food plan	Medical Outcome Study Short Form 36 questionnaire; University of Rhode Island Change Assessment questionnaire; Depression Anxiety Stress Scale questionnaire
Measuring the effects of caffeine and L-theanine on cognitive performance: a protocol for self-directed, mobile N-of-1 studies (NCT04056650)	Ongoing	Healthy volunteers	Repeated counterbalanced (ABBA or BAAB) N-of-1	Linear model with factors for treatment and block	Caffeine; Caffeine + L-theanine	Cognitive function via: 1) Remote Associates Test; 2) Stroop Test; 3) Trail Making Test
Tracking snacking in real time: time to look at individualized patterns of behavior	Published	healthy participants	observational N-of-1	Exploratory analysis; intra-class correlation coefficients	None	Consumption of high-calorie snack foods
The Diabetes Remission Clinical Trial (DiRECT): protocol for a cluster-randomized trial	Complete but with analysis ongoing	Patients with T2DM	Interventional and observational (subset of a large, cluster randomized trial)	Not stated for N-of-1 component	Low-calorie food replacement diet versus usual diabetes and obesity management	Adherence to dietary prescription as revealed by Ecological Momentary Assessment.

Adapted from Potter T, Vieira R, de Roos B. Adv Nutr. 2021;12:579-589.

N-of-1 Nutrition trials

Ma Y, Fu Y, Tian Y, Gou W, Miao Z, Yang M, Ordovás JM, Zheng JS. Individual Postprandial Glycemic Responses to Diet in n-of-1 Trials: Westlake N-of-1 Trials for Macronutrient Intake (WE-MACNUTR). J Nutr. 2021 Jul 13:nxab227. doi: 10.1093/jn/nxab227. Epub ahead of print. PMID: 34255080.



Men's and women's target energy intake was 2300 and 1900 kcal per day, respectively.

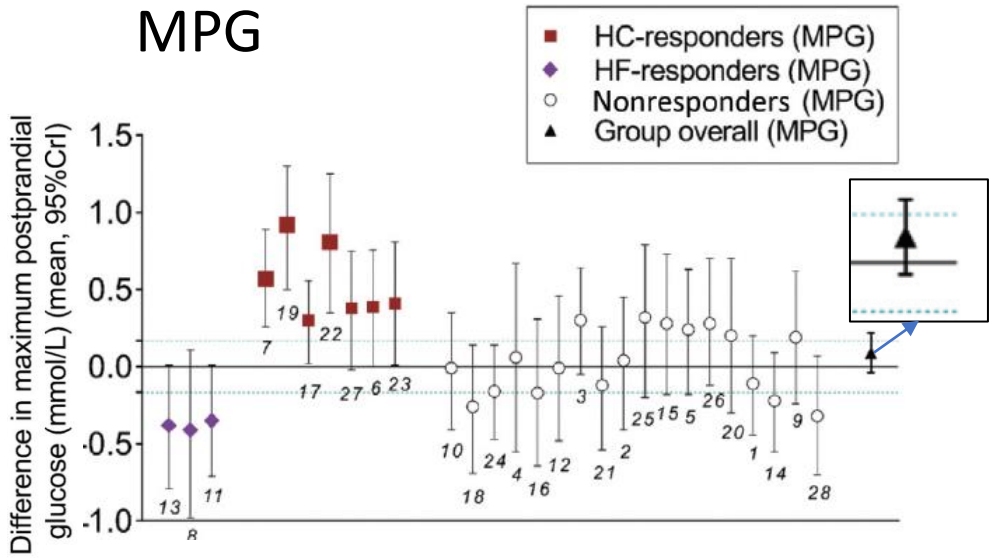
The HFLC diet consisted of 3 days of **70%E from fat, 15%E from protein, and 15%E from carbohydrate**, whereas the other 3-d diet consisted of **60%E from fat, 15%E from protein, and 25%E from carbohydrate**.

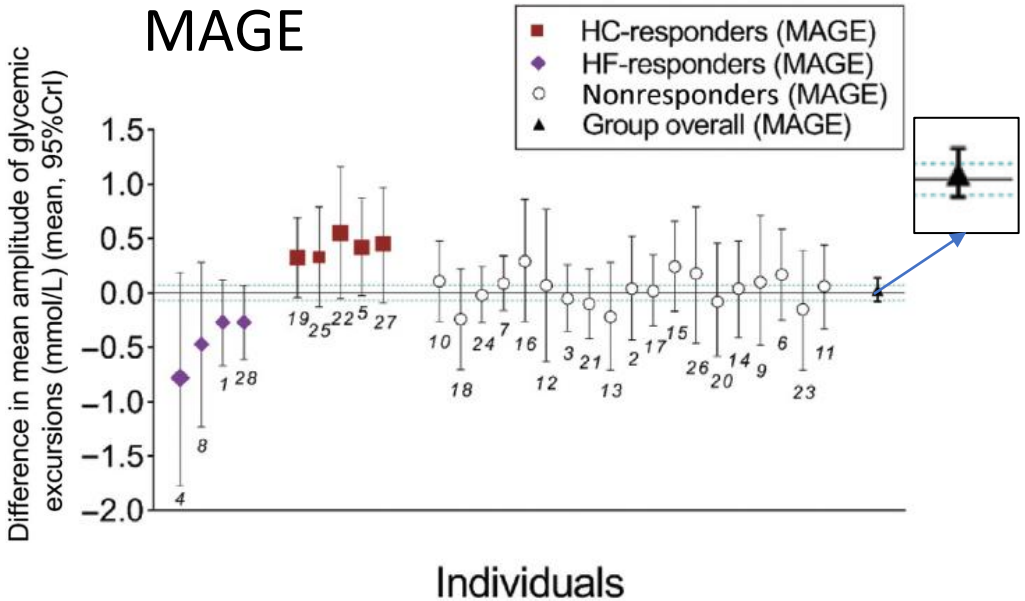
The LF-HC diet comprised a 3-d diet consisting of **20%E from fat, 15%E from protein, and 65%E from carbohydrate**, whereas the other 3-d diet consisted of **10%E from fat, 15%E from protein, and 75%E from carbohydrate**.

The diet for the 6-d wash-out period consisted of **30% from fat, 15% from protein, and 55% from carbohydrate**

WE-MACNUTR Main Results

maximum postprandial glucose (MPG), mean amplitude of glycemic excursions (MAGE), and AUC24 between intervention periods of LF-HC and HF-LC diets.





	Posterior probability (%)					
	Difference of MPG (mmol/L)		Difference of MAGE (mmol/L)		Difference of AUC_{24} (mmol/L·h)	
Participant *No.*	<−0.167	>0.167	<−0.072	>0.072	<−13.889	>13.889
1[2]	35.4	4.50	83.2	4.37	0	0
2	17.3	28.6	33.0	43.7	0	0
3	0.567	78.5	43.4	21.7	0	0.133
4[2]	22.7	37.2	91.6	4.47	0	0
5[3]	2.73	63.7	1.70	94.0	0.133	0
6[3]	0.367	87.9	12.6	66.5	0	0
7[3]	0	99.2	9.87	55.5	0	44.8
8[2]	81.0	1.60	83.8	8.60	3.07	0
9	5.17	53.9	28.5	52.9	0	0
10	20.7	17.7	17.2	56.5	0	0
11[2]	83.9	0.267	25.1	45.9	1.47	0
12	26.6	23.5	35.1	48.4	0	0
13[2]	85.1	0.167	72.6	13.1	0.0333	0
14	63.0	0.600	32.5	43.0	0	0
15	2.73	69.4	6.87	78.3	0	0
16	51.7	7.90	10.5	77.5	24.8	0
17[3]	0.0667	83.5	30.2	37.7	0	0
18	67.8	1.77	76.1	10.1	0	0
19[3]	0	100	1.77	91.5	0	61.8
20	7.20	55.6	51.8	27.6	0	0
21	41.6	7.53	58.1	16.0	0	0
22[3]	0	99.5	2.20	94.2	0	4.17
23[3]	0.400	88.7	60.3	21.5	0	22.9
24	47.7	1.70	34.7	23.1	0	0
25[3]	3.13	73.0	4.43	86.4	0	0
26	1.33	70.7	19.6	64.3	0	0
27[3]	0.433	86.3	3.03	91.7	0	0
28[2]	77.5	0.800	86.8	2.67	2.87	0

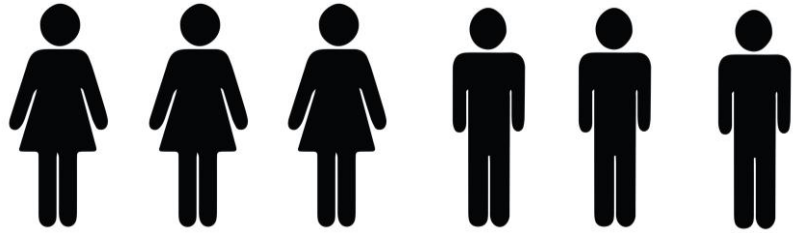
[1]Values are individual posterior probabilities (applied Bayesian analysis) of reaching a clinically meaningful difference in postprandial blood glucose between LF-HC and HF-LC diets, $n = 28$. This table reveals the posterior probability of the difference of outcomes between LF-HC and HF-LC periods reaching a clinically meaningful effect (MPG, 0.167 mmol/L; MAGE, 0.072 mmol/L; AUC_{24}, 13.889 mmol/L). HF-LC, high-fat, low-carbohydrate; LF-HC, low-fat, high-carbohydrate; MAGE, mean amplitude of glycemic excursions; MPG, postprandial maximum glucose.

[2]HF-responders for MPG or MAGE.

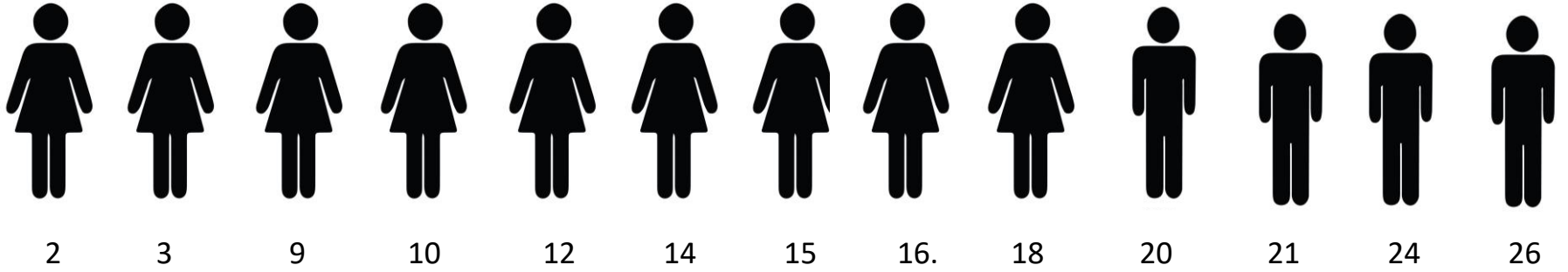
[3]HC-responders for MPG or MAGE.

HF/LC and LF/HC diets: Responders and non-responders from the n-of-1 trial. Escaping the tyranny of mean

5 6 7 17 19 22 23 25 27



1 4 8 11 13 28

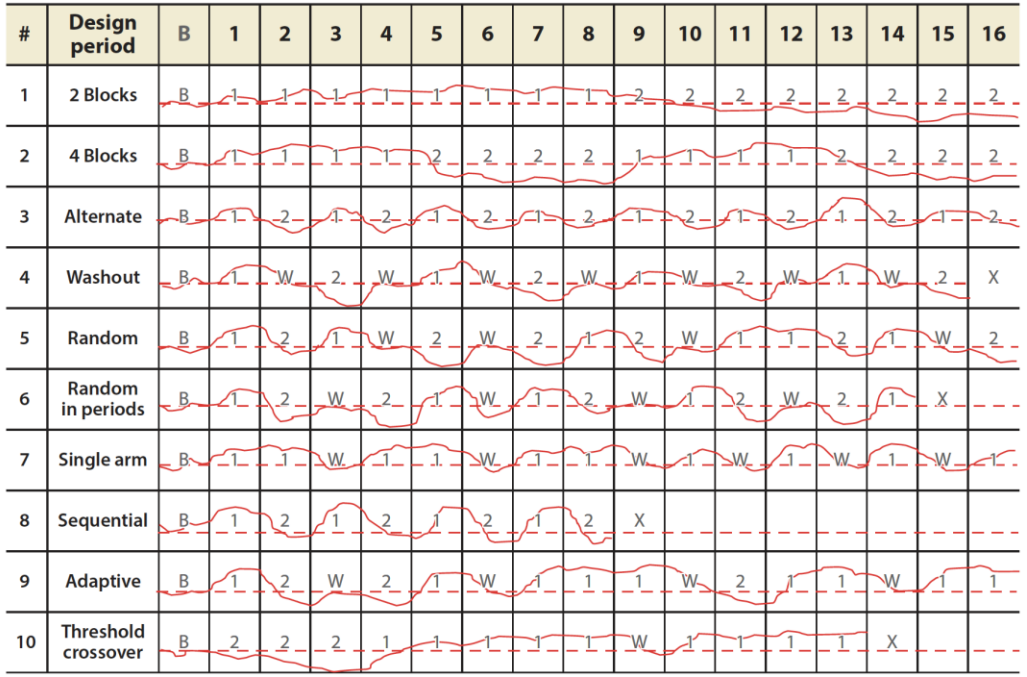


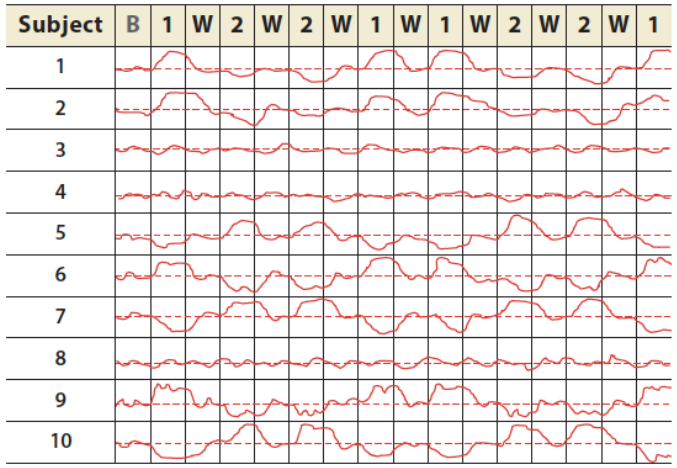
2 3 9 10 12 14 15 16. 18 20 21 24 26

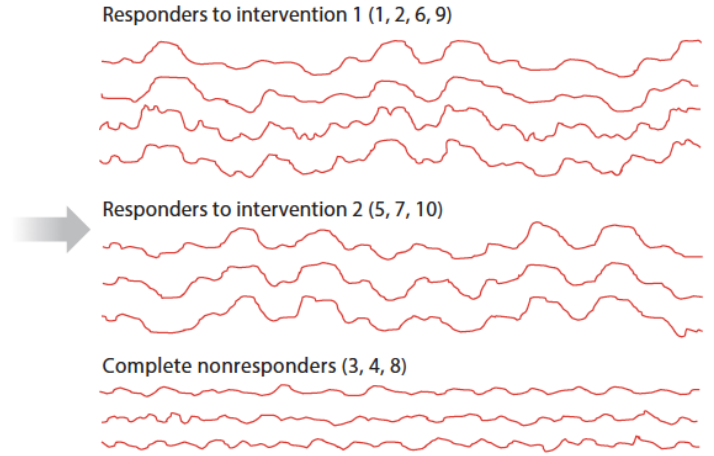
N-of-1 trials
aiming to answer nutritional questions one person at a time

Application of N-of-1 studies to nutrition research: considerations and challenges

- N-of-1 study designs involve the **repeated measurement** of a health outcome or behavior on an individual level.
- **Observational designs** can be used to monitor a participant's usual health or behavior in a naturalistic setting, with repeated measurements conducted in real time using an Ecological Momentary Assessment.
- **Interventional designs** can introduce a dietary or behavioral intervention with predictors and outcomes of interest measured repeatedly either during or after 1 or more intervention and control periods.
- Both types of design involve monitoring of a participant over time, and often employ **Ecological Momentary Assessment (EMA)** to obtain repeated measurements of health markers, behaviors, or attitudes.







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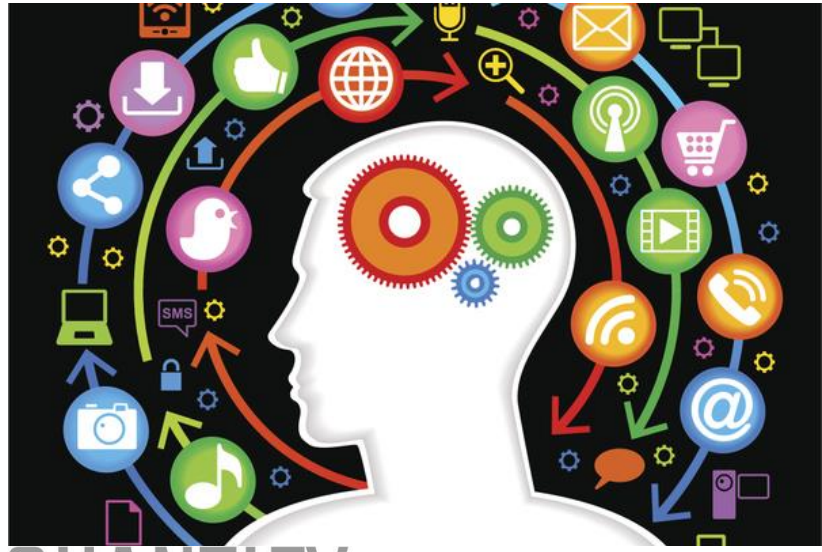
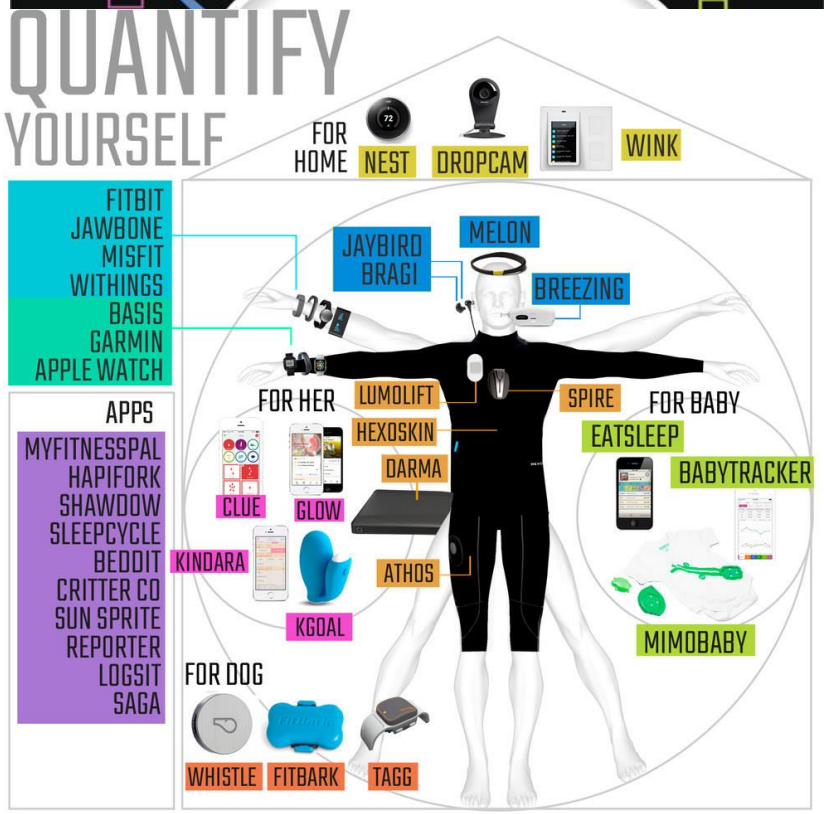
Application of N-of-1 studies to nutrition research: considerations and challenges

- A growing number of disease, behavioral, exposure markers can be measured outside of the clinic, with **self-reported data** delivered via electronic devices. Therefore, it is now easier than ever to generate large amounts of data on an individual level that could be particularly useful for application to **precision nutrition** studies.
- This may help to reveal novel associations between participant characteristics and health outcomes, with **repeated measures** providing power and precision to accurately: determine:
  - Determine an individual's **health status**
  - Predict **responses**
  - Recommend specific dietary/behavioral **recommendations**
- Although including repeated measurements of variable factors over the course of an N-of-1 study **increases the participant burden**, these measurements can also serve to **retain participant interest** if the researcher is able to build a profile on the participant that can be shared with them during or at the end of the study.





L²d Laboratory for Living Devices

Ph Precision health

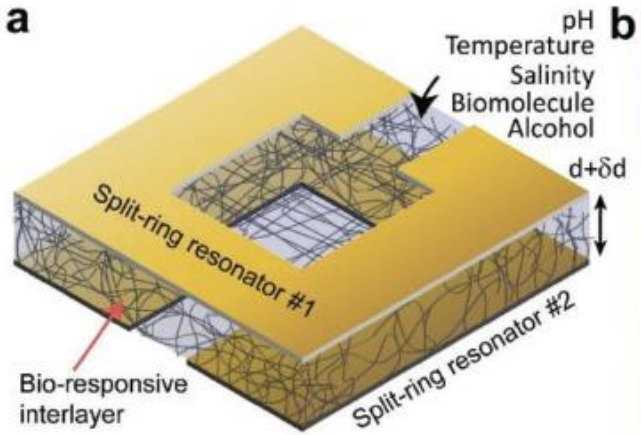
COMMUNICATION

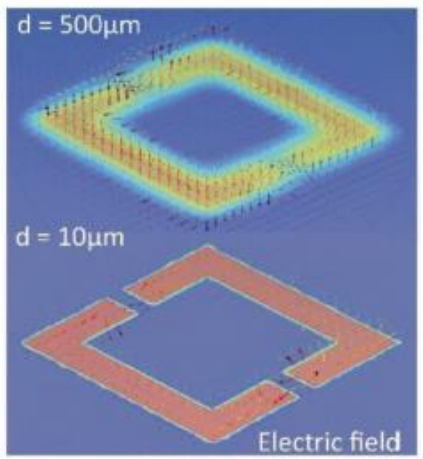
Wearable Sensors

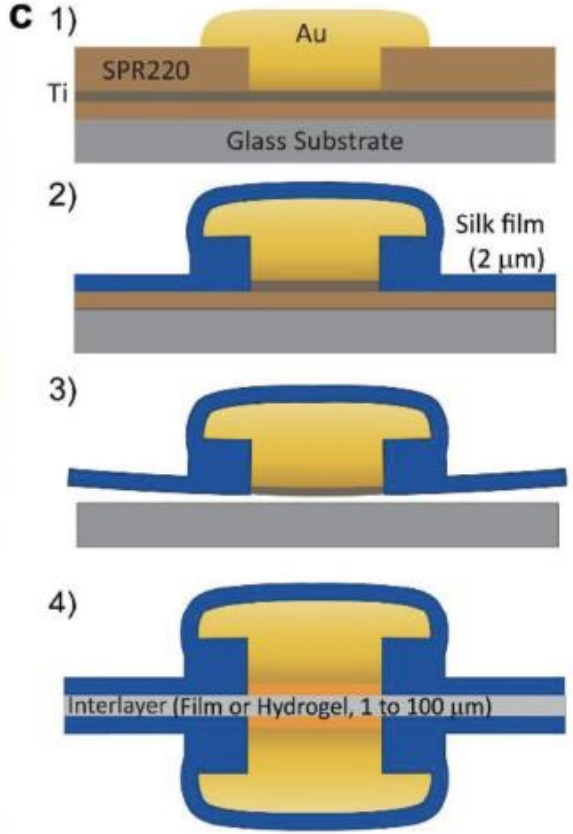
ADVANCED MATERIALS

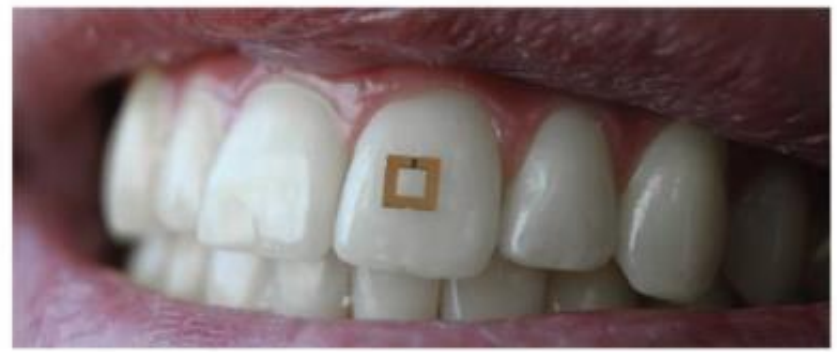
www.advmat.de

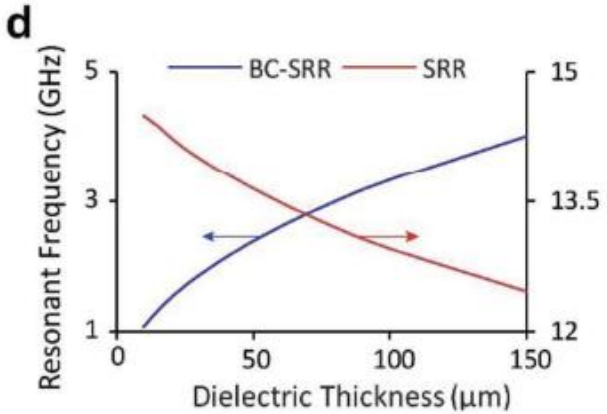
Functional, RF-Trilayer Sensors for Tooth-Mounted, Wireless Monitoring of the Oral Cavity and Food Consumption

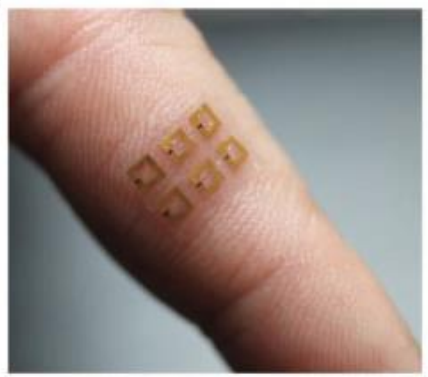


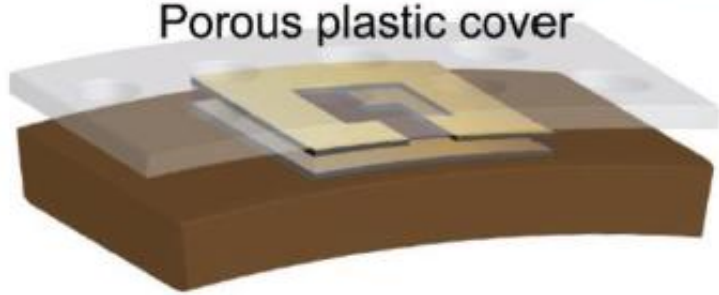




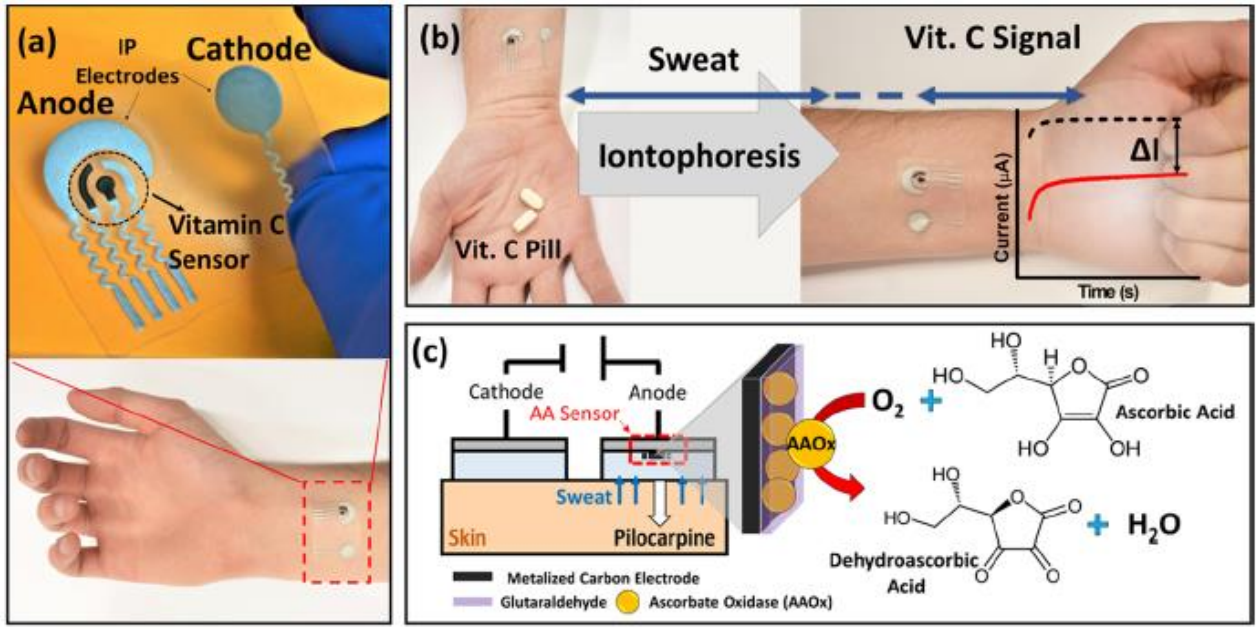




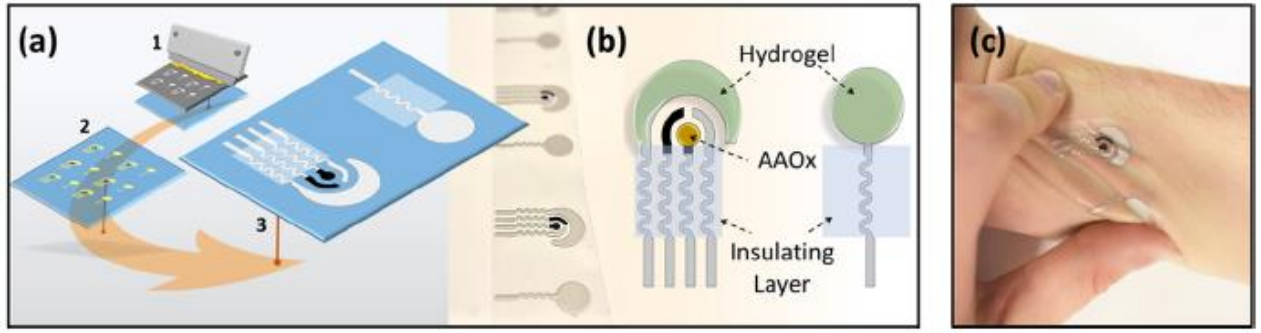




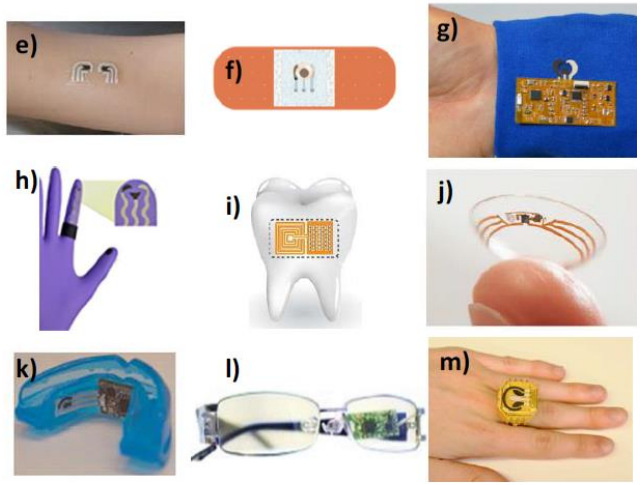
Epidermal Enzymatic Biosensors for Sweat Vitamin C: Toward Personalized Nutrition



Ascorbic acid (vitamin C) determination in stimulated sweat.
(a) Electrode design for simultaneous sweat stimulation and detection.
(b) Protocol used for the biosensing of ascorbic acid.
(c) Schematic of the localized sweat stimulation using IP pilocarpine delivery and of the enzymatic reaction for detecting ascorbic acid on a metalized Rh-carbon printed electrode.



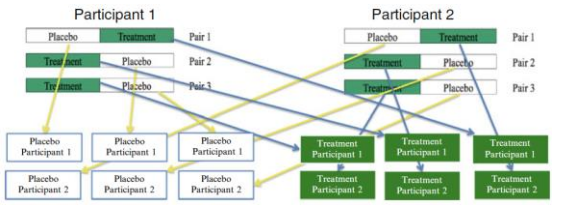
(a) Fabrication of the vitamin C biosensor.
(b) Schematic showing the location of the hydrogel and the enzyme layer.
(c) Image of the epidermal sensor under mechanical (twisting) strain.

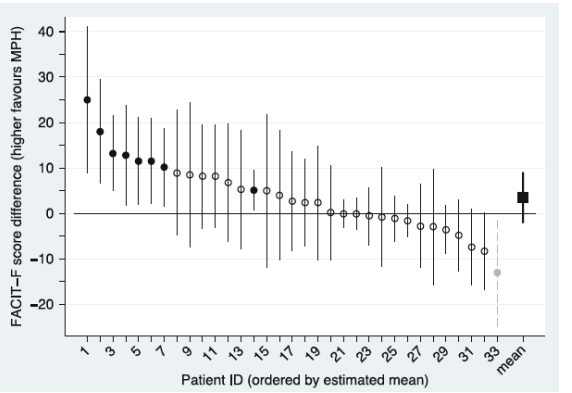


SPEs printed on flower (a), skull (b), panda bear (c), and marijuana (d) shapes. SPEs fabricated on a temporary tattoo (e), bendable bandage (f), textile substrate (g), glove (h), water-soluble silk thin-film substrates (transferred to tooth enamel) (i), contact lens (j), or incorporated in a mouthguard (k), eyeglasess (l) or ring (m).

Application of N-of-1 studies to nutrition research: considerations and **challenges**

- From <u>Funders/reviewers</u>
  - Proposals for N-of-1 trials can face some **criticism**. There may be an attempt to understand the study from the perspective of a group-level trial, including concerns that there is a lack of statistical power owing to the low number of participants (or a single participant) under study.
  - It should be explained/emphasized that **statistical power** is achieved from the number of measurements taken on an individual level.
  - Funders or institutes may not see the **utility** in conducting trials on an individual level or may believe it is not worth the amount of effort and resources required to examine a small number of participants.
  - Such criticisms can also affect the **interpretation** of results from an N-of-1 trial. However, individualized measures or single N-of-1 studies can help to identify variable factors that may affect health or behavior that could otherwise be overlooked, as analysis of time-course data can reveal associations which would be missed if fewer measurements were taken.
  - To investigate whether such associations are useful for other, similar patients, **aggregation** of sets of N-of-1 studies can be useful, particularly if a goal is to determine whether 1 intervention is superior to another.





Application of N-of-1 studies to nutrition research: considerations and **challenges**

From Health Professionals

- The average health **professional's toolbox** doesn't include the necessary statistical hammers and nails to do a meaningful analysis. "In clinical practice, health professionals use evidence and experience to generate a list of treatment options. Patients and health professionals move down the list based on trial and error."
- N-of-1 trials require health professionals to squeeze another process and script into their **already-packed days**. "Clinicians must explain the idea to their patients, see them regularly throughout the trial, and evaluate the results jointly at the trial's end. … Taking the time to sell an unfamiliar concept—let alone follow through on the logistics—might be untenable."

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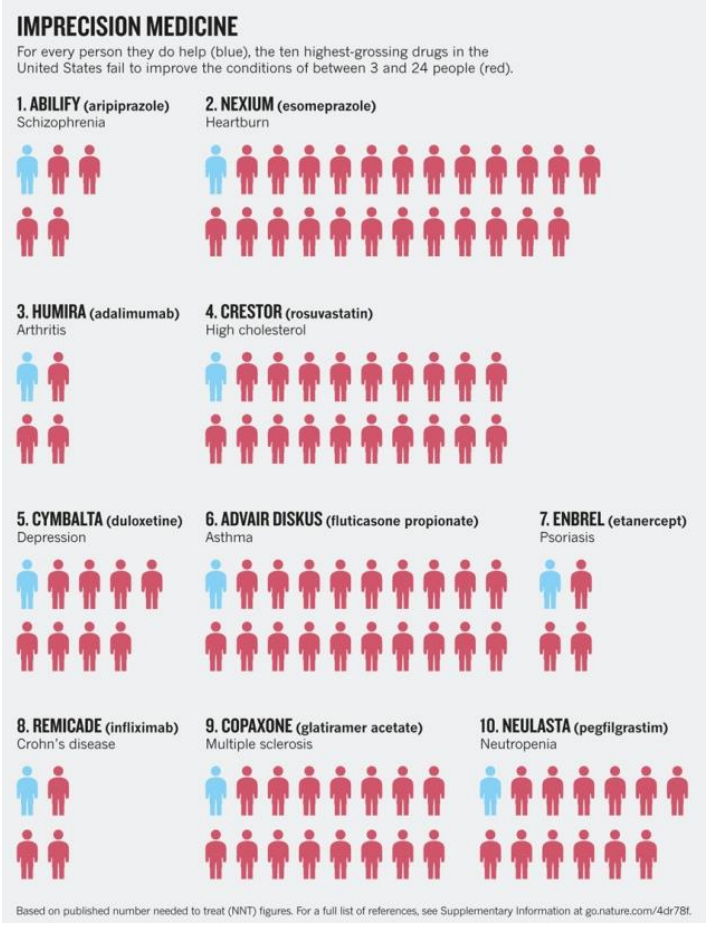
From Health Professionals (continued)

- Gordon Guyatt explains **resistance** to the n-of-1 innovation: "[Physicians] tend to have ways that they've learned to **operate where they're comfortable**, and it's hard to move outside these ways. It's hard to do something extra that's going to take time, and energy, and initiative."
- Nicholas J. Schork argues that **N-of-1 trials are long overdue:**
  - "Every day, millions of people are taking medications that will not help them,"
  - "The top ten highest-grossing drugs in the United States help between 1 in 25 and 1 in 4 of the people who take them. For some drugs, such as statins — routinely used to lower cholesterol — as few as 1 in 50 may benefit. There are even drugs that are harmful to certain ethnic groups because of the bias towards white Western participants in classical clinical trials."
  - "Physicians are having to become more acutely aware of the **unique circumstance of each patient** — something most people have long called for."
- These statements equally apply to **Precision Nutrition.**



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Imprecision nutrition? Different simultaneous continuous glucose monitors provide discordant meal rankings for incremental postprandial glucose in subjects without diabetes

Rebecca Howard, Juen Guo, and Kevin D Hall

National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD

Nutrition Research Limitations: Comparison traditional RCT vs. n-of-1 trial

Current Methods Used in Nutrition Research Limitations (particularly related to nutrition research)	RCT	n-of-1
Not being able to blind the interventions	⊖	⊖
Randomization does not guarantee balanced **covariate profiles** for any one trial	⊖	✓
Evidence suggesting equipoise between two (or more) interventions is often flawed, possibly leading to unethical trials, especially in an era of biomarker-enhanced personalized trials	⊖	✓
Not being able to recruit enough people for a RCT	⊖	✓⊖
Inability to recruit adequate number of participants with broad race/ethnicity distribution	⊖	✓⊖
Differences in habitual dietary patterns at onset of intervention and lack of incorporation of nutritional status in the design of the trial.	⊖	✓
Not being able to run the RCT for a long enough period of time (e.g., years) to observe the outcome of interest	⊖	⊖
The use of washout periods, especially with crossover trials, is not consistent, but may be necessary.	⊖	✓
Standard multi-arm RCTs don't explicitly take into consideration the value of rules dictating who should be provided the different treatments being tested, but instead randomize patients to trial arms. Such trials do not test the value of ways of assigning different treatments to subjects based on their, e.g., biomarkers	⊖	✓
Not being able to account for unique molecular, physiologic, environmental, socioecological determinants/factors and behavioral profiles of each individual when results are presented in aggregate and therefore not being able to make individualized or subgroup recommendations	⊖	✓
statistical bias due to confounding or omitted variables	⊖	✓
The "average effect" concept behind most population-based RCTs is vague and likely compromising insight into how to effectively deploy a new intervention	⊖	✓
Not being able to separate/understand all of the different pathways affecting an outcome	⊖	⊖✓
Focus on a single primary endpoint can complicate understanding of the broader and more systemic effects of an intervention	⊖	✓
Not able to extend a RCT/study to different populations, locations, circumstances, etc.	⊖	⊖✓
Unable to test for effectiveness in real world situation	⊖	✓
Low adherence	⊖	✓⊖
Focus on single nutrients – often leads to null results – we eat foods not single nutrients and there are established nutrient/nutrient interactions that need to be taken into consideration.	⊖	✓
The inter-individual background variation is often relatively large compared with the intra-individual effect of the intervention -	⊖	✓
Some argue that motivation and expectation are important factors in any behavioral intervention, including one focused on diet. Hence changes occur in the placebo group as well – need to think creatively about this and move away from a medical approach for diet	⊖	✓

In Conclusion