Effectiveness of Whey Protein Supplement in Resistance Trained Individuals

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Abstract

Athletes and recreationally resistance-trained individuals often use protein supplements in an attempt to maximize their training gains and performance. Because of the high bioavailability and solubility and its higher proportion of essential amino acids including Leucine, whey protein extract has been proposed as the best optimal form of protein for strength and power athletes. The objective of this review is to examine the current evidence for the efficacy of whey protein containing supplements to optimize strength training adaptation and outcomes for regular resistance training practitioners.

A limited numbers of studies have reported positive effects of whey protein containing supplements (including those with carbohydrate and creatine) for optimizing the anabolic responses and adaptations process in resistance-trained individuals.

In order to promote a more anabolic environment and maximize muscle protein synthesis along the day, an eating pattern behavior involving frequent meals (every 3 to 5 h) containing 17 to 20 g of high quality protein (200 to 250 mg/kg) providing 8 to 10 g of EAA (90 to 110 mg/kg) and about 2 g of Leucine (20 to 25 mg/kg) have been recommended. Special attention should be given to the preworkout hours where the ingestion of whey proteins combined with carbohydrates, creatine monohydrate (0.1 g/kg/d) and other proteins sources such as casein before, during and after workout have been shown to improve training adaptations and enhance the recovery process. However, when considering that the training conditions (workout volume, organization, number of exercises) used in the available studies are substantially different than what athletes actually perform. Optimal whey protein supplementation protocols need to specifically be based on the regular resistance training workout organization and would probably need to consider other doses and timing strategies than what is currently recommended.

Keywords: Multinutrient; Supplementation; Strength and power athletes; Amino acid; Leucine; casein; Sport-nutrition

Abbreviations: LBM: Lean Body Mass; AA: Amino Acid; PDCAAS: Protein Digestibility Corrected Amino Acid Score; DIAAS: Digestible Indispensable Amino Acid Score; EAA: Essential Amino Acid; BCAA: Branched Chain Amino Acid

Introduction

The protein requirements for athletic populations have been the subject of extensive scientific debate. During the last decade the notion that athletes as well generally active population require greater protein intake than the currently RDA recommendation of 0.8 g/kg of body weight per day in healthy adults has become well accepted [1,2]. In addition, high protein diets have also become quite popular in general population as part of many weight reduction programs [3,4]. Differences in protein requirements for different types of athletes (i.e., endurance, strength/power) and non-athletes are well documented [5-8]. Many studies have investigated the effects of protein obtained from different sources such as milk eggs soy rice or bovine colostrum as well as the technique used to manufacture the protein supplements or the methodology recommended for their ingestion [9-15]. In fact, these factors can be very relevant when the protein supplements are ingested with the scope to optimize training adaptations [16].

Although some studies have shown greater strength, muscular hypertrophy and/or Lean Body Mass (LBM) gains when resistance training is combined with whey protein alone or in conjunction with carbohydrates other protein sources enriched with amino acids or creatine; there are still some inconsistencies regarding the most appropriate effective doses and ingestion-timing strategy for optimizing resistance training induced adaptations and outcomes (strength gain or muscle mass accretion) [17-21].

As the focus of this review is on the effects of whey protein supplementation on both resistance training performances and their outcomes in trained individuals, we have focused on reviewing the evidence that have used whey protein preparation alone or combined with other protein sources or amino acid derivatives for optimizing resistance training responses for regular athletes and recreationally resistance training individuals. Thus, after describing the main characteristics of whey protein supplements, manufacture process, quality and Amino Acid (AA) profile; we will analyze the documented effects of whey protein containing supplements for resistance-trained individuals. In addition some practical recommendations about the most appropriate serving sizes and feeding distribution strategies will be provided based on current level of evidences.

Protein Supplements

Most of protein extract are obtained from milk, eggs, bovine colostrums, soy and to less extend from bean, wheat or rice. The quality of these extracts may vary depending on the type and quality...
of the manufacturing process. A range of processed milk proteins (concentrates, isolates, hydrolyzates or peptides) has been used for different purposes including change in metabolic health in young or elderly people or training optimization outcomes in untrained recreationally active or well trained athletes [19,22-26].

**Milk composition: Protein sub-fractions manufacturing and main differences**

Whole milk is approximately 87% water, with the remaining 13% as solids. The 13% solids are composed of 30% fat, 37% lactose, 27% protein, and 6% minerals. Of the 27% of milk that is protein, 20% is whey protein and 80% is casein protein [27]. During the process of making cheese, whey protein is separated out into a transparent liquid fraction that has shown to be a good source of high quality protein currently used to produce protein powders and many other products.

Milk protein concentrate, produced by ultrafiltration of skimmed milk, contains both casein and whey proteins in similar proportions to whole milk, but the total amount of protein, lactose and mineral content may vary between different formulations [14].

The casein fraction comprises α-casein, β-casein, and κ-casein whilst whey contain multiple globular proteins including β-lactoglobulin (50 to 55%), α-lactalbumin (20 to 25%) bovine serum albumin (5 to 10%) lactoferrin (1 to 2%) immunoglobulins (10 to 15%) lactoperoxidase enzymes (~0.5%) and glycomacropeptides (10 to 15%), enzymes and growth factors. All of these components have been associated with the potential to improve general health outcomes and disease prevention reduce immunosuppression attenuate muscular inflammatory related damage optimize recovery process and resistance training outcomes in athletes and enhance metabolic health in general population [19-22,28-30].

Whey fraction of the milk is particularly rich in essential amino acid, vitamins and mineral [14]. Protein concentrates obtained from whey are produced by coagulation of milk with the enzyme rennet or acid, resulting in separation of curds and whey, further ultrafiltration and drying produces whey protein concentrates containing ~25-80% protein [31]. Additional processing can produce whey protein isolates containing >90% protein with very low amounts of lactose and lipids [28]. Selective elution, also known as ion exchange chromatography, is the process most often used to produce whey protein isolates [14]. Additional hydrolysis with enzymes or acids provides a way to breakdown the structure of protein containing in whey [22]. As the acids hydrolysis process would also lower the quality and biological value of protein, the addition of proteolytic enzymes, followed by purification procedures is greatly preferred [32]. The main advantage of high quality whey protein is that it exchanges hydrolyzates preparations would be the rapid uptake and availability of AA and a possible strong insulinotropic effect that would elicit a fast and powerful stimulus on muscle protein synthesis during the 3 h postprandial period [33,34]. This advantageous postprandial effect of whey protein contributes to numerous anabolic responses following resistance training and its supplementation has been recently shown to have superior cardio-protective effects on resting condition while after training the ingestion of one single dose of whey protein has been proposed as to be more effective to stimulate anabolic responses in resistance trained individuals [5,25].

Amino acid profile of whey and casein protein supplements

Based on human AA requirements, digestibility and their bioavailability, similar to eggs protein, both whey and casein protein have been classified as high quality proteins [31]. Considering both the protein digestibility corrected AA score (PDCAAS) and the recently developed digestible indispensable AA score (DIAAS), casein and whey protein fractions contain higher amount and proportion of essential amino acids (EAA) compared to other vegetable protein sources such as soy or rice [42]. Nevertheless, in addition to their different digestive patterns, some special physiological effects have also been attributed to their particular AA composition [43]. Figure 1 compares the AA compositions of the whey and casein protein fractions [24].

Data from Figure 1 indicates that when the same amount of casein or whey protein are consumed, whey provide more than 3% of EAA and higher amount of Cysteine, and Leucine. Cysteine is thought to play a key role in the regulation of whole body protein metabolism,
tyrosine in addition to support phenylalanine levels, is essential AAs L-cysteine and taurine, all of which play vital roles in human health glutathione, the polyamines spermine and spermidine, as well as the donor reactions for the synthesis of epinephrine, creatine, melatonin, muscle [30]. Methionine is an essential AA associated with the methyl and modulating muscle contraction, glucose and AAs uptake by the important regulatory role by increasing blood flow to the muscles L-arginine is the main physiological precursor of nitric oxide that plays ammonia detoxification, hormone secretion, and immune modulation. L-Arginine, L-Taurine, L-Methionine and L-Tyrosine [14,31]. For could be fortified with the addition of peptides of L-Glutamine, of the whey protein extract, a high quality based whey protein product metabolic functions, in order to maximize the nutritional proprieties of the whey protein extract, a high quality based whey protein product could be fortified with the addition of peptides of L-Glutamine, L-Arginine, L-Taurine, L-Methionine and L-Tyrosine [14,31]. For example, glutamine accounts for 30-35% of the amino acid nitrogen in the plasma being the most abundant amino acid in blood. L-glutamine fulfills a number of biochemical needs. It operates as a nitrogen shuttle, taking up excess ammonia and forming urea. It can act as a powerful anticitaboic agent contributing for the production of other AAs, glucose, nucleotides, protein, and glutathione [42]. Arginine is a semi-essential AA, involved in numerous areas of human biochemistry, including ammonia detoxification, hormone secretion, and immune modulation. L-arginine is the main physiological precursor of nitric oxide that plays an important regulatory role by increasing blood flow to the muscles and modulating muscle contraction, glucose and AAs uptake by the muscle [30]. Methionine is an essential AA associated with the methyl donor reactions for the synthesis of epinephrine, creatine, melatonin, glutathione, the polyamines spermere and spermidine, as well as the AAs L-cysteine and taurine, all of which play vital roles in human health [48]. Tyrosine in addition to support phenylalanine levels, is essential for the production of catecholamine neurotransmitters, including dopamine, norepinephrine, and epinephrine. Tyrosine is the precursor for the hormones thyrara and triiodothyronine by the thyroid gland and grow hormone by the pituitary [49]. There is some evidence that supplementation with L-tyrosine can lead to an increase in dopamine that in turn may lead to an improvement in mood state [50]. Although, each protein source has unique attributes that may convey specific nutritional advantages compared to the others (general health benefits, body weight management, gaining lean body mass, enhancing muscle recovery after exercise or stimulating muscular protein synthesis) the degree by which each source of protein can stimulate these process are related to different factors involving digestion rates, divergent AA profiles, pattern of ingestion and the metabolic state (rest or exercise) related to the timing of ingestion [39,40].

Documented Effects of Whey Protein Supplement to Optimize Resistance-Training Responses

Whey protein extract, because of its increased bioavailability and solubility, and its higher percentage content of EAA including BCAAs, specifically Leucine, has been proposed as the best optimal form of protein for strength and power athletes [17,45,51]. Although several studies have analysed the effect of whey protein supplementation on resistance training performance and outcomes only some of them have focused on resistance trained individuals [12,17,23,24,26,38,52,53].

Cribb et al., [45] reported higher significant improvements in LBM, strength and loss of body fat in a group of recreational bodybuilders who supplemented their diet with the addition of 1.5 g /kg/d of hydrolyzed whey protein isolate compared to other group who ingested the same amount of casein during a supervised 10-week resistance training program. The 1.5 g/kg/d supplement dose was divided into smaller equal servings and consumed throughout the day (e.g. breakfast, lunch, immediately after training, and dinner).

Kraemer et al., [53], analysed the effects of 14 days supplementation period in 10 resistance trained males who were exposed to three different supplementation protocols: 20 g of whey protein isolate, 20 g of soy protein isolate or 20 g of carbohydrates. Each participant completed all three conditions separated by a two weeks wash out period. Lower cortisol and higher total testosterone levels were measured from 5 to 30 min after performing a squat protocol involving 6 sets of 10
repetitions at 80% 1RM followed by the whey protein supplementation period compared to when participants were exposed to soy or placebo. In addition, whey supplementation might also attenuate the normal increase of cortisol observed after carbohydrate condition.

Tang et al. [17] reported positive effects of 10 g of whey protein combined with 21 g of fructose, to induce a 2-fold rise in muscle protein synthesis in young well resistance trained men, over 4 h, after performing 4 sets of 8–10 repetitions at 80% 1RM for the leg press and leg extension exercises.

As whey protein contains about 50% of EAA, 10 g of whey high quality extract would provide circa 5 g of EAA [24]. If we consider that 20 g of whole egg protein, containing 8.6 g of EAA including 2 g or Leucine, have been necessary to maximally stimulate muscle and albumin protein synthesis after a lower-body resistance exercise in young men it may be possible that even when 10 g of whey protein were sufficient to elicit significant increases of muscle protein synthesis, a higher dose (≈17 g) would be necessary to achieve the optimal maximal response beyond which a marked stimulation of whole-body AA oxidation with no further increase in muscle protein synthesis would occur in young resistance trained males [10].

High-quality proteins extract, such as whey, stimulate muscle protein synthesis in proportion to the amount of EAA and specifically Leucine administered per serving [38]. Norton and Wilson suggest that optimal protein intake per meal should be based on the amount of leucine provided by the source of protein consumed [54]. Thus, the presence of the relative amount of the EAA including Leucine is of cornerstone importance for determining the amount of protein to be ingested in every meal. In fact when the amount of EAA and Leucine are equated, the effects on muscle protein synthesis and training outcomes seems to be similar regardless of which protein source is consumed [12,54]. Joy et al. [12], observed no differences of ingesting either 48 g of isolate protein extracted from rice or whey during 8 weeks of daily ondulated periodized resistance training program on short term recovery (Ratings of perceived recovery, soreness, and readiness) or training-induced adaptations (increase lean body mass, muscle mass, strength and power and decrease fat mass) in 24 resistance trained young males. Even when the absence of control group did not allow the authors to conclude how beneficial whey or casein protein supplementation would be compared to resistance training alone, based on previous investigations it could be possible to say that regardless of the source, between 17 to 20 g of high quality protein (200 to 250 mg/kg) providing 8 to 10 g of EAA (90 to 110 mg/kg) and about 2 g of Leucine (20 to 25 mg/kg) would be necessary to elicit an optimal muscle protein synthesis response in young strength trained individuals after performing a typical resistance training workout [10,17]. As a consequence, when ingested others plant-based protein extracts such as soy or rice containing less proportion of EAA and leucine, in order to obtain similar results, higher doses should be administered. However, larger protein ingestion entail also higher caloric intake greater digestion time (slower release of AA to the periphery), and increased AA oxidation and ureagenesis because more proportion of AA would be converted to urea rather than used for protein synthesis [5].

Summarizing the previous information, even when the need of EAA and Leucine could be satisfied from different protein sources, whey protein extract seems to be more efficient, on a gram per gram basis.

Whey protein combined with carbohydrates, other proteins sources, amino acid and/or creatine

Added carbohydrates to whey protein extract have been shown to enhance cellular hydration, glycogen resynthesis and reduce muscle protein breakdown which then magnifies the increase in net protein balance and/or lessen the immunosuppressive effects of exercise compared to the ingestion of whey protein or AA alone [8,55,56]. These positive effects are in part related to a more powerful insulin secretion [54]. With regard to resistance training, the ingestion of 40 g of whey protein with 120 of sucrose, honey powder or maltodextrine (1:3 protein to carbohydrate ratio) following a typical resistance training workout involving 9 exercises of 3 sets per 10 rep at 70% 1RM, did significantly influence glucose and insulin responses without significantly altering markers of anabolism, catabolism or immunity during the first two hours of recovery [18].

Multinutrients supplements containing whey protein, carbohydrates, amino acids, and other natural compounds such a creatine may optimize recovery time and training induced adaptation [19-21,57-59].

Kerksick et al. [20] reported significant greater increases in fat free mass in 36 resistance-trained men after combining a 10 weeks strength training program with the ingestion of 40 g whey plus 8 g casein and 2 g of high quality protein (200 to 250 mg/kg) providing 8 to 10 g of EAA including 2 g of Leucine or 40 g of whey enriched with 3 g of BCAAs, 5 g of glutamine and 2 g of carbohydrates. Supplements where ingested within 2 hours after workout during training days or at the morning on non-training days. Authors concluded that co-ingestion of blend high quality whey and casein proteins mix may be an effective means to promote higher improvements on body composition in resistance-trained individuals compared to the ingestion of only one protein source.

As stated previously whey and casein possess different patterns of amino acid release, which has been shown to greatly affect the extracellular amino acid concentrations and the resulting levels of protein synthesis and breakdown [60]. Differences in digestion rates would cause a more sustained increase in blood amino acid levels, resulting in greater muscle amino acid uptake [39]. Casein tends to inhibit protein catabolism when consumed at rest and has a slight, but relatively long increase in protein synthesis. The pattern of amino acid delivery with casein appears to lead to better leucine balance (net protein state) than whey at rest [5]. Conversely, after a typical resistance training workout whey protein have shown to stimulates a greater acute (0-3 h post-exercise) rise in muscle protein synthesis compared with the same amount of casein and is still highly effective for stimulating muscle protein synthesis over 3-5 h post-exercise [60-62]. Thus, possibly when both types of proteins sources are combined, in addition to a more favorable AA muscle blood-flow after workout, a higher aminoacidemia would be sustained along the day. As the stimulation of protein synthesis may be largely depend on the availability of the extracellular concentration of AA, for resistance trained practitioners, the co-ingestion of fast (whey) and slow (casein) high quality protein extract would elicit a more powerful anabolic and effective stimulus compared to only one source.

Cribb et al. [21] examined the effects of 11 weeks of resistance training program combined with whey protein and/or creatine monohydrate (both separately and in combination) on body composition, muscle strength, and muscular hypertrophy in a group of recreational male bodybuilders. Participants were dived in 4 groups: whey protein; whey protein plus creatine monohydrate; carbohydrates and carbohydrates plus creatine monohydrate. Supplementation with whey protein, whey protein plus creatine monohydrates and carbohydrates plus creatine monohydrate resulted in greater hypertrophy responses and strength.
gains compared to the ingestion of only carbohydrates. Additionally, the consumption of a creatine monohydrate in conjunction with whey proteins or carbohydrates provides similar benefits that in addition were greater than those obtained from carbohydrates or whey protein alone. In the line with the previous investigation, Cooper et al. [59] reported greater possibilities for improving 1 RM bench press (78%) and upper and lower body muscular endurance (49%) when a group of resistance trained males who performed a 12 weeks resistance-training program ingested 2 serving of 60 g per day of a multinutrient containing 21 g of carbohydrate, 30 g of whey protein, 4.7 g of fat, 5.1 g of creatine, 5.1 g of glutamine and 1.5 g HMB instead of a carbohydrate-placebo.

Summarizing the previous information, in addition to the synergic effects of adding carbohydrates, in order to optimize the positive effects of whey protein for improving resistance training adaptation, combining other protein sources such as casein and specifically the addition of creatine monohydrate would be appropriate. On the other hand, the additional benefits on resistance training outcomes resulted from the addition of other AA or proteins derivatives such as glutamine, BCAAs or HMB remain to be elucidated.

**Whey protein supplementation protocol: the nutrient-timing strategy**

Unfortunately there is no conclusive evidence about what time point and dose are optimal for the ingestion of protein or amino acid-carbohydrate supplement for optimizing muscular adaptations to exercise. Many studies have provided positive results for ingesting high quality protein before, during, immediately after or several hours (1 to 3-5 h) post exercise for promoting increases in protein synthesis [58]. With regard to the most convenient nutrition-time-protocol for improving resistance training outcomes, Cribb and Hayes reported significantly greater increases in lean body mass, strength and hypertrophy after 10 weeks of resistance training in a group of trained males who ingested a multinutrient contained glucose, whey isolate, creatine monohydrate and fat just before and after workout compared to another group which consumed the same dose before breakfast and just prior retiring to bed [26].

The optimal feeding strategy to maximally enhance recovery and anabolic response to resistance exercises should also consider the serving size and feeding distribution before, during and after workout. West et al., [25] observed greater muscular protein synthesis for 5 h of recovery in 8 recreationally trained males when consuming a 25 g single-bolus-dose of whey protein after performing 8 sets of 8-10 repetitions of a bilateral leg extension compared to the same amount of proteins ingested as repeated, small pulsed drinks (10 doses of 2.5 g of whey protein every 20 min). In another similar study, Burke et al. [63] observed different patterns of aminoacidemia in 12 resistance trained males who ingested 25 g of whey protein enriched with 5 g of Leucine or placebo before performing 1 set x 8-10 repetition at 80% 1RM in a single leg extension exercise. Supplements were administered in a single-bolus-dose (45 min before) or as a series of 15 small-pulsed drinks, (of approximately 33 mL each) starting 45 min before and ended 2 h after completed the training. Although similar increases in muscle protein synthesis (near 250% respect to placebo) were observed over 5 h after exercise, in the early phase of recovery the pulsed strategy produced higher AA and insulin concentration.

If we assume that between 17 to 20 g of high quality protein providing 8 to 10 g of EAA are necessary to maximally stimulates muscle protein synthesis after exercises, considering that a typical resistance training workout enhance muscle protein synthesis for at least 24 h; in order to maximize the anabolic effects of feeding it should be necessary to maintain a frequent protein ingestion, not only during the periworkout period but also along the day [64,65]. Because muscle protein synthesis becomes refractory to persistent aminoacidemia in order to maintain an optimal protein synthesis stimulation, between 5 to 6 meals providing 17 to 20 g of high quality protein should be recommended [54]. In line with this rationale, Areta et al., [66] examined the effects of three different protein feeding distributions over a 12 h recovery period in 24 resistance trained male who performed 4 sets of 10 repetitions at 80% 1RM with 3 min rest between sets followed by the ingestion of 80 g of whey protein isolate. Participants were divided in three groups: Pulse (ingested 8 servings of 10 g every 1.5 h); Intermediate (ingested 4 servings of 20 g every 4 h) and bolus (ingested 2 servings of 40 g every 6 h). Results showed higher myofibrillar protein synthesis stimulation for the intermediate protocol (4 intakes of 20 g) compared to both pulse (8 servings of 10 g) and bolus (2 servings of 40 g). Possibly 10 g of whey protein is not enough and resulted in some suboptimal stimulation of the protein synthesis. In contrast, larger but less frequent servings of 40 g, even if should be appropriate to maximally stimulate muscle protein synthesis, represent a suboptimal ingestion pattern due to irreversible amino acid oxidation of the excess ingested protein and possible the lack of anabolic stimulus between intakes (>5 h).

In summary as the cell membrane is highly sensitive to the internal and external (blood) rise of AA concentration, frequent meals (every 3 to 5 h) containing 17 to 20 g or high quality protein would potentiate and maintain a long lasting and optimal muscular protein synthesis stimulus. Additionally, with the aim to maximize the anabolic response of resistance training workout a higher protein intake (bolus) should be provided immediately after training. Figure 2 depicts the theoretical optimal protein-feeding pattern protocol starting 3 h before (small intakes =10 g) and ending 6 h after training (higher intakes = 20 g).

It is important to note that even when high quality whey protein extract would be an appropriate option for ingesting during training and just after workout others protein sources including those coming from regular food such as eggs, meet, cheese etc can be used along the different daily meals [67].

![Figure 2: Proposed ideal protocol of protein feeding for the periworkout hours.](image-url)
Contraindications and Precautions Related to Whey Protein Extract Ingestion

To date, no severe adverse reactions have been reported following administration of whey protein extracts. Although some individuals, who are hypersensitive to milk proteins, can experience allergy symptoms including possible serious ones (in this case is strongly recommended to avoid whey proteins containing products). Other dairy-sensitive individuals are lactose-intolerant. Most whey proteins are processed to remove lactose and finished whey products only contain trace amounts. De-lactosed whey, produced from crystallizing a majority of the lactose out and recovering the remaining whey, is appropriate for lactose-intolerant individuals [28].

Conclusions and Final Recommendations

The optimal protocol for administering a supplement containing high quality whey protein still needs to be determined. However, in order to stimulate a more powerful anabolic response, frequent meals (every 3 to 5 h) containing 17 to 20 g of high quality protein (200 to 250 mg/kg) providing 8 to 10 g of EAA (90 to 110 mg/kg) and about 2 g of Leucine (20 to 25 mg/kg) seems appropriate for eliciting optimal muscular responses and adaptations in resistance training practitioners.

Co-ingestion of whey protein and carbohydrates in 1:3 ratio would helps glycogen restoration and inhibits protein catabolism, creating synergistic effects with the EAA, particularly leucine, for optimizing recovery process after resistance training workout [68].

Special attention should be given to the perworkout hours where the ingestion of small amount of whey protein (=10 g) under a pulse like protocol should begin 3 h before and continue through the training session. A higher protein intake (=20 g) would be recommended immediately post workout and possibly at 3 and 6 h after training. In addition, combined whey with others protein sources such as casein and specifically with creatine monohydrate (0.1 g/kg/d), would be appropriate for improving recovery process and the anabolic responses induced by resistance training in athletes [69].

Most available studies have analysed the results based on serial biopsies of only one muscle and limited training conditions (low volume and number of exercises) that are substantially different from what athletes actually perform.

Future research may explore the responses of regular resistance training practitioners under most realistic training condition, using others timing strategies and possibly higher proteins doses than what is currently being recommended.

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